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(57) Abstract

The present invention relates, in general, to a method of modulating physiological and pathological processes and, in particular, to a method of modulating cellular levels of oxidants and thereby processes in which such oxidants are a participant. The invention also relates to compounds and compositions suitable for use in such methods.

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SUBSTITUTED PORPHYRINS

TECHNICAL FIELD

The present invention relates, in general, to a method of modulating physiological and pathological processes and, in particular, to a method of modulating cellular levels of oxidants and thereby processes in which such oxidants are a participant. The invention also relates to compounds and compositions suitable for use in such methods.

BACKGROUND

Oxidants are produced as part of the normal metabolism of all cells but also are an important component of the pathogenesis of many disease processes. Reactive oxygen species, for example, are critical elements of the pathogenesis of diseases of the lung, the central nervous system and skeletal muscle. Oxygen free radicals also play a role in modulating the effects of nitric oxide (NO·). In this context, they contribute to the pathogenesis of vascular disorders, inflammatory diseases and the aging process.

A critical balance of defensive enzymes against oxidants is required to maintain normal cell and organ function. Superoxide dismutases (SODs) are a family of metalloenzymes that catalyze the intra- and extracellular conversion of O_2^- into H_2O_2 plus O_2, and represent the first line of defense against the detrimental effects of superoxide radicals. Mammals produce three distinct SODs. One is a dimeric copper- and zinc-containing enzyme (CuZn SOD) found in the cytosol of all cells. A second is a tetrameric manganese-containing SOD (Mn SOD) found within mitochondria, and the third is a tetrameric, glycosylated, copper- and zinc-containing enzyme (EC-SOD) found in the extracellular fluids and bound to the extracellular matrix. Several other important antioxidant enzymes are known to exist within cells, including

catalase and glutathione peroxidase. While extracellular fluids and the extracellular matrix contain only small amounts of these enzymes, other extracellular antioxidants are also known to be present, including radical scavengers and inhibitors of lipid peroxidation, such as ascorbic acid, uric acid, and α-tocopherol (Halliwell et al, Arch. Biochem. Biophys. 280:1 (1990)).

The present invention relates generally to low molecular weight porphyrin compounds suitable for use in modulating intra- and extracellular processes in which superoxide radicals, or other oxidants such as hydrogen peroxide or peroxynitrite, are a participant. The compounds and methods of the invention find application in various physiologic and pathologic processes in which oxidative stress plays a role.

SUMMARY OF THE INVENTION

The present invention relates to a method of modulating intra- or extracellular levels of oxidants such as superoxide radicals, hydrogen peroxide, peroxynitrite, lipid peroxides, hydroxyl radicals and thiyl radicals. More particularly, the invention relates to a method of modulating normal or pathological processes involving superoxide radicals, hydrogen peroxide, nitric oxide or peroxynitrite using low molecular weight antioxidants, and to methine (ie, *meso*) substituted porphyrins suitable for use in such a method.

Objects and advantages of the present invention will be clear from the description that follows.

BRIEF DESCRIPTION OF THE DRAWING

Figure 1 shows the structures of certain compounds of the invention. The SOD activity values were determined using the method of McCord and Fridovich, J. Biol. Chem. 244:6049 (1969). The TBARS values were obtained as follows:

Homogenates

Frozen adult Sprague-Dawley rat brains, livers and mouse lungs (Pel-Freez, Rogers, AR) were homogenized with a polytron (Turrax T25, Germany) in 5 volumes of ice cold 50 mM potassium phosphate at pH 7.4: Homogenate protein concentration was determined with the Coomassie Plus protein assay (Pierce, Rockford, IL) using bovine serum albumin as a standard. The homogenate volume was adjusted with buffer to give a final protein concentration of 10mg/ml and frozen as aliquots at -80°C.

Oxidation of homogenates

Microfuge tubes (1.5 ml) containing 0.2 ml of homogenate (0.2 mg protein) and various concentrations of antioxidant were incubated at 37°C for 15 minutes. Oxidation of the rat brain homogenate was initiated by the addition of 0.1 ml of a freshly prepared stock anaerobic solution containing ferrous chloride (0.25 mM) and ascorbate (1 mM). Samples were placed in a shaking water bath at 37°C for 30 minutes (final volume 1 ml). The reactions were stopped by the addition of 0.1 μL of a stock butylated hydroxytoluene (60 mM) solution in ethanol.

Lipid peroxidation measurement

The concentration of thiobarbituric acid reactive species (TBARS) in rat brain homogenates was used as a index of lipid peroxidation. Malondialdehyde standards were obtained by adding 8.2 μL of 1,1,3,3-tetramethoxypropane in 10 ml of 0.01 N HCl and mixing for 10 minutes at room temperature. This stock was further diluted in water to give standards that ranged from 0.25 to 25 μM. Samples or standards (200 μL) were acidified with 200 μL of 0.2 M stock of phosphoric acid in 1.5 ml locking microfuge tubes. The color reaction was initiated by the addition of 25 μL of a stock thiobarbituric acid solution (0.11M) that was mixed and then placed in a 90°C heating block for 30 minutes. TBARS were extracted with 0.5 ml of n-butanol by vortexing for 3 minutes and chilling on ice for 1 minute. The samples were then centrifuged at 12,000 x g

for 3 minutes and a 150 μL aliquot of the n-butanol phase was placed in each well of a 96-well plate and read at 535 nm in a Thermomax platereader (Molecular Devices, Sunnydale, CA) at 25°C. Sample absorbances were converted to MDA equivalences (μM) by extrapolation from the MDA standard curve. None of the antioxidants at concentrations employed in these studies affected the reaction of MDA standards with thiobarbituric acid.

Statistical analyses

Data were presented as their means $\pm$ SE. The inhibitory concentration of antioxidants that decreased the degree of lipid peroxidation by 50% (IC_{50}) and respective 95% confidence intervals (CI) were determined by fitting a sigmoidal curve with variable slope to the data (Prizm, GraphPad, San Diego, CA). (See also Braughler et al, J. Biol. Chem. 262:10438 (1987); Kikugawa et al, Anal. Biochem. 202:249 (1992).)

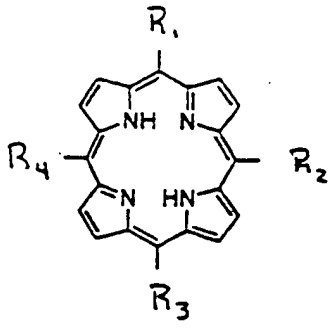
DETAILED DESCRIPTION OF THE INVENTION

The present invention relates to methods of protecting against the deleterious effects of oxidants, particularly, superoxide radicals, hydrogen peroxide and peroxynitrite, and to methods of preventing and treating diseases and disorders that involve or result from oxidant stress. The invention also relates methods of modulating biological processes involving oxidants, including superoxide radicals, hydrogen peroxide, nitric oxide and peroxynitrite. The invention further relates to compounds and compositions, including low molecular weight antioxidants (eg mimetics of scavengers of reactive oxygen species, including mimetics of SODs, catalases and peroxidases) and formulations thereof, suitable for use in such methods.

Mimetics of scavengers of reactive oxygen species appropriate for use in the present methods include methine (ie *meso*) substituted porphines, or pharmaceutically acceptable salts thereof (eg chloride or bromide salts). The invention includes both metal-free and metal-bound porphines. In the case of metal-bound porphines, manganic derivatives of methine (*meso*) substituted

porphines are preferred, however, metals other than manganese such as iron (II or III), copper (I or II), cobalt (II or III), or nickel (I or II), can also be used. It will be appreciated that the metal selected can have various valence states, for example, manganese II, III or V can be used. Zn (II) can also be used even though it does not undergo a valence change and therefore will not directly scavenge superoxide. The choice of the metal can affect selectivity of the oxygen species that is scavenged. Iron-bound porphines, for example, can be used to scavenge NO· while manganese-bound porphines scavenge less well.

The mimetics of the present invention are of the Formula I:



I

or pharmaceutically acceptable salt thereof

wherein:

R_1 and R_3 are, independently:

$-CO_2C_{1-4}$ alkyl; or

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3;

R_2 is:

-H

$-C_{1-4}$alkyl

-COOH

$-CO_2C_{1-4}$ alkyl,

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

$-CON(CH_3)_2$, or

$-CX_3$, wherein X is halogen; and

R_4 is:

-H,

$-C_{1-4}$alkyl

-COOH,

$-CO_2C_{1-4}$ alkyl,

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

$-CON(CH_3)_2$, or

$-CX_3$, wherein X is halogen.

Preferably, R_1 and R_3 are, independently, $-CO_2C_{1-4}$alkyl (advantageously, $-CO_2C_{1-3}$alkyl) or $-CO_2CH_2CX_3$ (advantageously, where X = F), R_2 is -H, $-CO_2C_{1-3}$alkyl, $-CO_2CH_2CX_3$ (advantageously, where X = F), $-CON(CH_3)_2$ or CX_3 (advantageously, where X = F) and R_4 is -H, -COOH, $-CO_2C_{1-3}$alkyl, $-CON(CH_3)_2$ or $-CX_3$ (advantageously, where X = F).

More preferably, R_1 and R_3 are, independently, $-CO_2C_{1-3}$alkyl, R_2 is $-CO_2C_{1-3}$alkyl, $-CON(CH_3)_2$ or $-CX_3$ (advantageously, where X = F), and R_4 is -H, -COOH, $-CO_2C_{1-3}$alkyl, $-CON(CH_3)_2$ or $-CX_3$ (advantageously, where X = F).

Still more preferably, R_1 or R_3 is $-CO_2CH_3$, $-CO_2CH_2CH_3$, or $-CON(CH_3)_2$, R_2 is $-CO_2CH_3$, $-CO_2CH_2CH_3$, or CX_3 (advantageously, where X = F), and R_4 is -H, -COOH, $-CO_2CH_3$, $-CO_2CH_2CH_3$ or CX_3 (advantageously, where X = F).

Even more preferably, R_1, R_2 and R_3 are, independently, $-CO_2CH_3$ or $-CO_2CH_2CH_3$, and R_4 is -H, -COOH, $-CO_2CH_3$ or $-CO_2CH_2CH_3$ or $-CO_2CH_2CH_3$.

Most preferably, R_1, R_2, R_3 and R_4 are, independently, $-CO_2CH_3$ or $-CO_2CH_2CH_3$.

Specific examples of mimetics of the invention are shown in Figure 1, together with activity data.

In addition to the methine (*meso*) substituents described above, one or more of the pyrrole rings of the porphyrin of Formula I can be substituted at any or all beta carbons, ie: 2, 3, 7, 8, 12, 13, 17 or 18. Such substituents, designated P, can be hydrogen or an electron withdrawing group, for example, each P can, independently, be a NO_2 group, a halogen (eg Cl, Br or F), a nitrile group, a vinyl group, or a formyl group. Such substituents alter the redox potential of the porphyrin and thus enhance its ability to scavenge oxygen radicals. For example, there can be 1, 2, 3, 4, 5, 6, 7, or 8 halogen (eg Br) substituents (preferably, 1-4), the remaining P's advantageously being hydrogen. When P is formyl, it is preferred that there not be more than 2 (on non-adjacent carbons), more preferably, 1, the remaining P's preferably being hydrogen. When P is NO_2, it is preferred that there not be more than 4 (on non-adjacent carbons), more preferably, 1 or 2, the remaining P's being hydrogen.

Where isomers are possible, all such isomers of the herein described mimetics are within the scope of the invention.

Mimetics preferred for use in the present methods can be selected by assaying for SOD, catalase and/or peroxidase activity. Mimetics can also be screened for their ability to inhibit lipid peroxidation.

SOD activity can be monitored in the presence and absence of EDTA using the method of McCord and Fridovich (J. Biol. Chem. 244:6049 (1969)). The efficacy of a mimetic can also be determined by measuring the effect of the mimetic on the aerobic growth of a SOD null *E. coli* strain versus a parent strain. Specifically, parental *E. coli* (AB1157) and SOD null *E. coli*. (JI132) can be grown in M9 medium containing 0.2% casamino acids and 0.2% glucose at pH 7.0 and 37°C; growth can be monitored in terms of turbidity followed at 700 nm. This assay can be made more selective for SOD mimetics by omitting the branched chain, aromatic and sulphur-containing amino acids from the medium (glucose minimal medium (M9), plus 5 essential amino acids).

Efficacy of active mimetics can also be assessed by determining their ability to protect mammalian cells against methylviologen (paraquat)-induced

toxicity. Specifically, rat L2 cells grown as described below and seeded into 24 well dishes can be pre-incubated with various concentrations of the SOD mimetic and then incubated with a concentration of methylviologen previously shown to produce an LC_{75} in control L2 cells. Efficacy of the mimetic can be correlated with a decrease in the methylviologen-induced LDH release (St. Clair et al, FEBS Lett. 293:199 (1991)).

The efficacy of SOD mimetics can be tested *in vivo* with mouse and/or rat models using both aerosol administration and parenteral injection. For example, male Balb/c mice can be randomized into 4 groups of 8 mice each to form a standard 2X2 contingency statistical model. Animals can be treated with either paraquat (40 mg/kg, ip) or saline and treated with SOD mimetic or vehicle control. Lung injury can be assessed 48 hours after paraquat treatment by analysis of bronchoalveolar lavage fluid (BALF) damage parameters (LDH, protein and % PMN) as previously described (Hampson et al, Tox. Appl. Pharm. 98:206 (1989); Day et al, J. Pharm. Methods 24:1 (1990)). Lungs from 2 mice of each group can be instillation-fixed with 4% paraformaldehyde and processed for histopathology at the light microscopic level.

Catalase activity can be monitored by measuring absorbance at 240nm in the presence of hydrogen peroxide (see Beers and Sizer, J. Biol. Chem. 195:133 (1952)) or by measuring oxygen evolution with a Clark oxygen electrode (Del Rio et al, Anal. Biochem. 80:409 (1977)).

Peroxidase activity can be measured spectrophotometrically as previously described by Putter and Becker: Peroxidases. In: Methods of Enzymatic Analysis, H.U. Bergmeyer (ed.), Verlag Chemie, Weinheim, pp. 286-292 (1983). Aconitase activity can be measured as described by Gardner and Fridovich (J. Biol. Chem. 266:19328 (1991)). The selective, reversible and SOD-sensitive inactivation of aconitase by known O^{-}_2 generators can be used as a marker of intracellular O^{-}_2 generation. Thus, suitable mimetics can be selected by assaying for the ability to protect aconitase activity.

The ability of mimetics to inhibit lipid peroxidation can be assessed as described by Ohkawa et al (Anal. Biochem. 95:351 (1979)) and Yue et al (J. Pharmacol. Exp. Ther. 263:92 (1992)). Iron and ascorbate can be used to initiate lipid peroxidation in tissue homogenates and the formation of thiobarbituric acid reactive species (TBARS) measured.

Active mimetics can be tested for toxicity in mammalian cell culture by measuring lactate dehydrogenase (LDH) release. Specifically, rat L2 cells (a lung Type II like cell (Kaighn and Douglas, J. Cell Biol. 59:160a (1973)) can be grown in Ham's F-12 medium with 10% fetal calf serum supplement at pH 7.4 and 37/C; cells can be seeded at equal densities in 24 well culture dishes and grown to approximately 90% confluence; SOD mimetics can be added to the cells at log doses (eg micromolar doses in minimal essential medium (MEM)) and incubated for 24 hours. Toxicity can be assessed by morphology and by measuring the release of the cytosolic injury marker, LDH (eg on a thermokinetic plate reader), as described by Vassault (In: Methods of Enzymatic Analysis, Bergmeyer (ed) pp. 118-26 (1983); oxidation of NADH is measured at 340 nm).

Synthesis of various mimetics suitable for use in the present method can be effected using art-recognized protocols (see, for example Sastry et al, Anal. Chem. 41:857 (1969), Pasternack et al, Biochem. 22:2406 (1983); Richards et al, Inorg. Chem. 35:1940 (1996) and U.S. Appln. No. 08/663,028, particularly the details therein relating to syntheses). Synthesis of a number of mimetics of the invention are set forth in the Examples that follow.

The mimetics of the present invention are suitable for use in a variety of methods. The compounds of Formula I, particularly the metal bound forms (advantageously, the manganese bound forms), are characterized by the ability to inhibit lipid peroxidation. Accordingly, these compounds are preferred for use in the treatment of diseases or disorders associated with elevated levels of lipid peroxidation. The compounds are further preferred for use in the treatment of

diseases or disorders mediated by oxidative stress. Inflammation diseases are an example.

The compounds of the invention (advantageously,metal bound forms thereof) can also be used in methods designed to regulate NO· levels by targeting the above-described porphines to strategic locations. NO· is an intercellular signal and, as such, NO· must traverse the extracellular matrix to exert its effects. NO·, however, is highly sensitive to inactivation mediated by O_2^- present in the extracellular spaces. The methine (*meso*) substituted porphyrins of the invention can increase bioavalability of NO· by preventing its degradation by O_2^- .

The present invention relates, in a further specific embodiment, to a method of inhibiting production of superoxide radicals. In this embodiment, the mimetics of the invention (particularly,metal bound forms thereof) are used to inhibit oxidases, such as xanthine oxidase, that are responsible for production of superoxide radicals. The ability of a mimetic to protect mammalian cells from xanthine/xanthine oxidase-induced injury can be assessed, for example, by growing rat L2 cells in 24-well dishes. Cells can be pre-incubated with various concentrations of a mimetic and then xanthine oxidase (XO) can be added to the culture along with xanthine (X). The appropriate amount of XO/X used in the study can be pre-determined for each cell line by performing a dose-response curve for injury. X/XO can be used in an amount that produces approximately an LC_{75} in the culture. Efficacy of the mimetic can be correlated with a decrease in XO/X-induced LDH release.

The mimetics of the invention (particularly, metal bound forms thereof) can also be used as catalytic scavengers of reactive oxygen species to protect against ischemia reperfusion injuries associated with myocardial infarction, stroke, acute head trauma, organ reperfusion following transplantation, bowel ischemia, hemorrhagic stock, pulmonary infarction, surgical occlusion of blood flow, and soft tissue injury. The mimetics (particularly, metal bound forms) can further be used to protect against skeletal muscle reperfusion injuries. The

mimetics (particularly, metal bound forms) can also be used to protect against damage to the eye due to sunlight (and to the skin) as well as glaucoma, and macular degeneration of the eye. Diseases of the bone are also amenable to treatment with the mimetics. Further, connective tissue disorders associated with defects in collagen synthesis or degradation can be expected to be susceptible to treatment with the present mimetics (particularly, metal bound forms), as should the generalized deficits of aging.

The mimetics of the invention (particularly, metal bound forms) can also be used as catalytic scavengers of reactive oxygen species to increase the very limited storage viability of transplanted hearts, kidneys, skin and other organs and tissues. The invention also provides methods of inhibiting damage due to autoxidation of substances resulting in the formation of O_2^- including food products, pharmaceuticals, stored blood, etc. To effect this end, the mimetics of the invention are added to food products, pharmaceuticals, stored blood and the like, in an amount sufficient to inhibit or prevent oxidation damage and thereby to inhibit or prevent the degradation associated with the autoxidation reactions. (For other uses of the mimetics of the invention, see USP 5,227,405). The amount of mimetic to be used in a particular treatment or to be associated with a particular substance can be determined by one skilled in the art.

The mimetics (particularly, metal bound forms) of the invention can further be used to scavenge hydrogen peroxide and thus protect against formation of the highly reactive hydroxyl radical by interfering with Fenton chemistry (Aruoma and Halliwell, Biochem. J. 241:273 (1987); Mello Filho et al, Biochem. J. 218:273 (1984); Rush and Bielski, J. Phys. Chem. 89:5062 (1985)). The mimetics (particularly, metal bound forms) of the invention can also be used to scavenge peroxynitrite, as demonstrated indirectly by inhibition of the oxidation of dihydrorhodamine 123 to rhodamine 123 and directly by accelerating peroxynitrite degradation by stop flow analysis.

Further examples of specific diseases/disorders appropriate for treatment using the mimetics of the present invention, advantageously, metal bound forms,

include diseases of the central nervous system (including AIDS dementia, stroke, amyotrophic lateral sclerosis (ALS), Parkinson's disease and Huntington's disease) and diseases of the musculature (including diaphramic diseases (eg respiratory fatigue in emphysema, bronchitis and cystic fibrosis), cardiac fatigue of congestive heart failure, muscle weakness syndromes associated with myopathies, ALS and multiple sclerosis). Many neurologic disorders (including stroke, Huntington's disease, Parkinson's disease, ALS, Alzheimer's and AIDS dementia) are associated with an over stimulation of the major subtype of glutamate receptor, the NMDA (or N-methyl-D-aspartate) subtype. On stimulation of the NMDA receptor, excessive neuronal calcium concentrations contribute to a series of membrane and cytoplasmic events leading to production of oxygen free radicals and nitric oxide (NO·). Interactions between oxygen free radicals and NO· have been shown to contribute to neuronal cell death. Well-established neuronal cortical culture models of NMDA-toxicity have been developed and used as the basis for drug development. In these same systems, the mimetics of the present invention inhibit NMDA induced injury. The formation of O^{-}_{2} radicals is an obligate step in the intracellular events culminating in excitotoxic death of cortical neurons and further demonstrate that the mimetics of the invention can be used to scavenge O^{-}_{2} radicals and thereby serve as protectants against excitotoxic injury.

The present invention also relates to methods of treating AIDS. The Nf Kappa B promoter is used by the HIV virus for replication. This promoter is used by the HIV virus for replication. This promoter is redox sensitive, therefore, an oxidant can regulate this process. This has been shown previously for two metalloporphyrins distinct from those of the present invention (Song et al, Antiviral Chem. and Chemother. 8:85 (1997)). The invention also relates to methods of treating arthritis, systemic hypertension, atherosclerosis, edema, septic shock, pulmonary hypertension, including primary pulmonary hypertension, impotence, infertility, endometriosis, premature uterine contractions, microbial infections, gout and in the treatment of Type II diabetes

mellitus. The mimetics of the invention (particularly, metal bound forms) can be used to ameliorate the toxic effects associated with endotoxin, for example, by preserving vascular tone and preventing multi-organ system damage.

As indicated above, inflammations, particularly inflammations of the lung, are amenable to treatment using the present mimetics (particularly, metal bound forms) (note particularly the inflammatory based disorders of asthma, ARDS including oxygen toxicity, pneumonia (especially AIDS-related pneumonia), cystic fibrosis, chronic sinusitis and autoimmune diseases (such as rheumatoid arthritis)). EC-SOD is localized in the interstitial spaces surrounding airways and vasculature smooth muscle cells. EC-SOD and O_2^- mediate the antiinflammatory - proinflammatory balance in the alveolar septum. NO· released by alveolar septal cells acts to suppress inflammation unless it reacts with O_2^- to form $ONOO^-$. By scavenging O_2^-, EC-SOD tips the balance in the alveolar septum against inflammation. Significant amounts of $ONOO^-$ will form only when EC-SOD is deficient or when there is greatly increased O_2^- release. Mimetics described herein can be used to protect against destruction caused by hyperoxia.

The invention further relates to methods of treating memory disorders. It is believed that nitric oxide is a neurotransmitter involved in long-term memory potentiation. Using an EC-SOD knocked-out mouse model (Carlsson et al, Proc. Natl. Acad. Sci. USA 92:6264 (1995)), it can be shown that learning impairment correlates with reduced superoxide scavenging in extracellular spaces of the brain. Reduced scavenging results in higher extracellular O^-_2 levels. O^-_2 is believed to react with nitric oxide thereby preventing or inhibiting nitric oxide-mediated neurotransmission and thus long-term memory potentiation. The mimetics of the invention, particularly, metal bound forms, can be used to treat dementias and memory/learning disorders.

The availability of the mimetics of the invention also makes possible studies of processes mediated by O_2^-, hydrogen peroxide, nitric oxide and peroxynitrite.

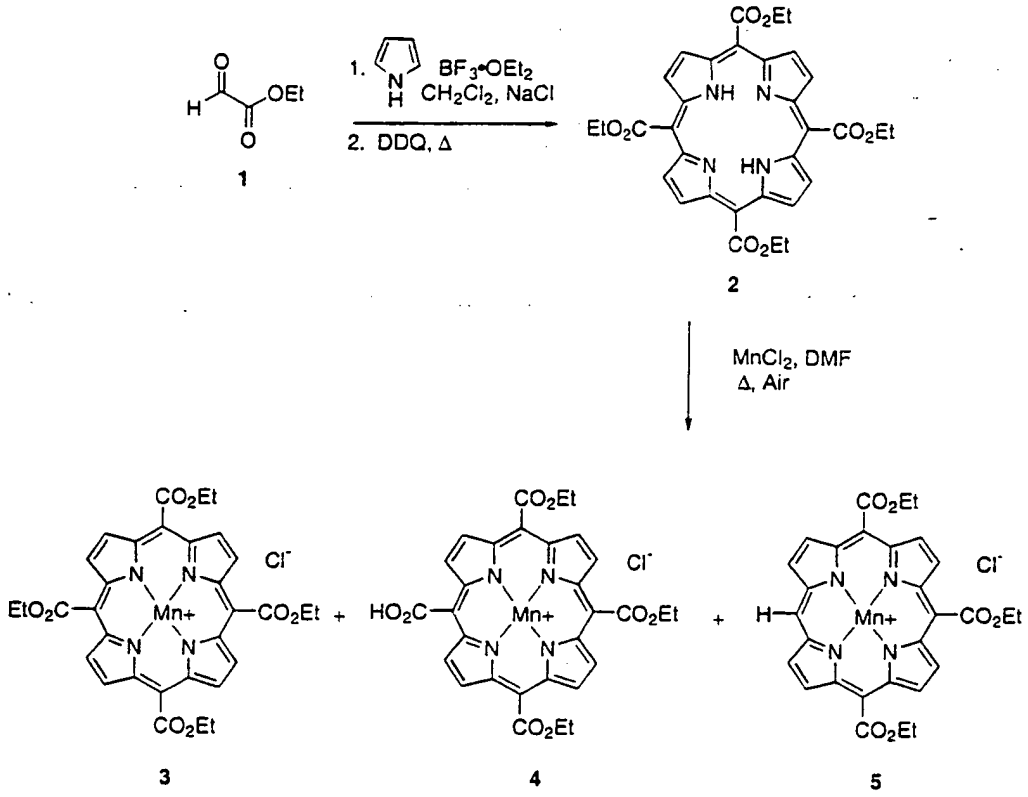
The mimetics described above, metal bound and metal free forms, can be formulated into pharmaceutical compositions suitable for use in the present methods. Such compositions include the active agent (mimetic) together with a pharmaceutically acceptable carrier, excipient or diluent. The composition can be present in dosage unit form for example, tablets, capsules or suppositories. The composition can also be in the form of a sterile solution suitable for injection or nebulization. Compositions can also be in a form suitable for opthalmic use. The invention also includes compositions formulated for topical administration, such compositions taking the form, for example, of a lotion, cream, gel or ointment. The concentration of active agent to be included in the composition can be selected based on the nature of the agent, the dosage regimen and the result sought.

The dosage of the composition of the invention to be administered can be determined without undue experimentation and will be dependent upon various factors including the nature of the active agent (including whether metal bound or metal free), the route of administration, the patient, and the result sought to be achieved. A suitable dosage of mimetic to be administered IV or topically can be expected to be in the range of about 0.01 to 50 mg/kg/day, preferably, 0.1 to 10 mg/kg/day. For aerosol administration, it is expected that doses will be in the range of 0.001 to 5.0 mg/kg/day, preferably, 0.01 to 1 mg/kg/day. Suitable doses of mimetics will vary, for example, with the mimetic and with the result sought.

Certain aspects of the present invention will be described in greater detail in the non-limiting Examples that follow.

EXAMPLE 1

I. [5,10,15,20-Tetrakis(ethoxycarbonyl)porphyrinato]manganese(III) Chloride (3), [5-Carboxy-10,15,20-tris(ethoxycarbonyl)porphyrinato]-manganese(III) Chloride (4) and [5,10,15-Tris(ethoxycarbonyl)-porphyrinato]manganese(III) Chloride (5).



1. 5,10,15,20-Tetrakis(ethoxycarbonyl)porphyrin (2).

In a foil covered, 22 L three-neck round-bottom flask equipped with a mechanical stirrer and a N_2 inlet was added consecutively, freshly distilled ethyl glyoxylate (Hook, J. M. *Synth. Commun.* **1984**, *14*, 83-87) (19.3 g, 189 mmol), CH_2Cl_2 (19 L), NaCl (1.1 g, 19 mmol) and pyrrole (13.1 mL, 189 mmol). The reaction mixture was stirred for 5-10 min then $BF_3•OEt_2$ (7.0 mL, 56 mmol) was added dropwise. After a stirring period of 1.25 h at room temperature, 2,3-

dichloro-5,6-dicyano-1,4-benzoquinone (DDQ, 32.2 g, 142 mmol) was added. The reaction mixture was stirred for an additional 2 h at room temperature, then clay (Clarion 550, 99 g) was added and the reaction mixture was stirred overnight. Filtration of the heterogeneous mixture through Celite, followed by evaporation of solvents provided a solid mixture which was then adsorbed onto silica gel (24 g). Repeated chromatographic purifications (5 batches; CH_2Cl_2 as eluent) on silica gel provided compound **4** (1.7 g; 6%) as a dark solid: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.33 (s, 2 H), 1.81 (t, 12 H), 5.11 (q, 8 H), 9.52 (s, 8 H); ^{13}C NMR (75 MHz, $CDCl_3$) δ 14.97, 63.67, 112.43, 131.64, 145.30, 170.68.

2. [5,10,15,20-Tetrakis(ethoxycarbonyl)porphyrinato]manganese(III) Chloride (3).

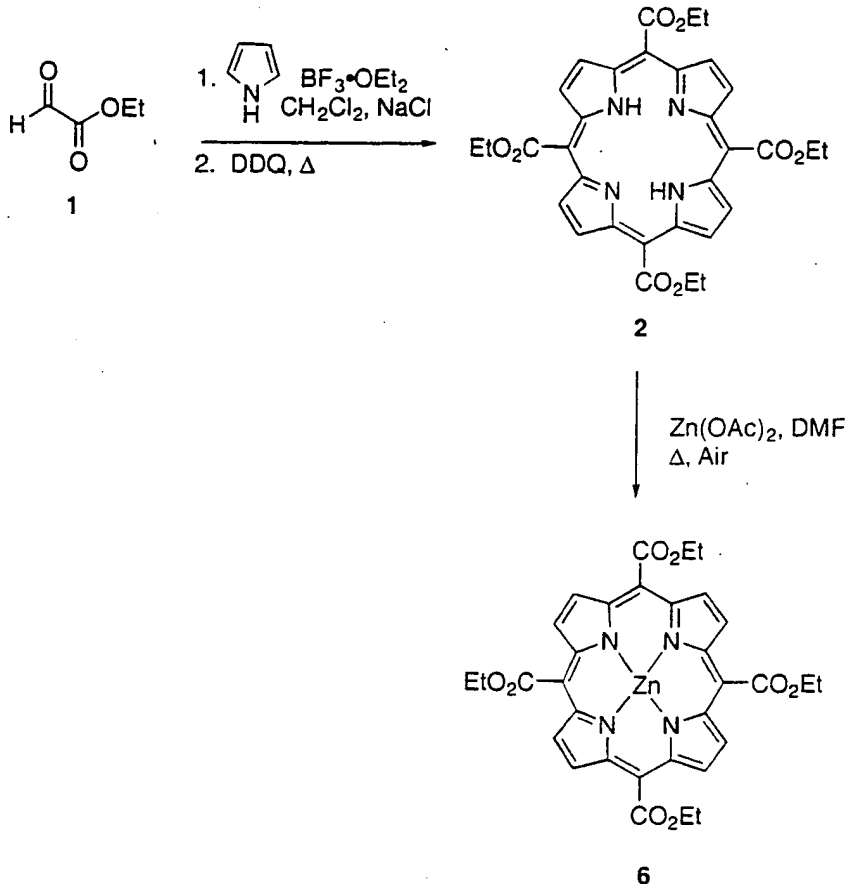
A solution of **2** (4.43 g, 7.4 mmol) and $MnCl_2$ (4.67 g, 37.1 mmol) in DMF (500 mL) was heated at 145 °C for 1-1.5 h then exposed to a stream of air. The reaction mixture was heated for an additional 2-3 h then allowed to cool to room temperature overnight and under a stream of air. Evaporation of the DMF provided a solid mixture which was adsorbed onto silica gel (24 g). Purification by column chromatography (6 batches; gradient elution 0→7% MeOH/CH_2Cl_2) provided porphyrin **3** (4.3 g; 84%) as a dark solid: mp > 300 °C; UV-vis λ_{max}= 456 nm, ε = 1.08 x 10^5 L/cm-mole; FAB MS m/z = 651 $[C_{32}H_{28}MnN_4O_8]^+$.

3. [5-Carboxy-10,15,20-tris(ethoxycarbonyl)porphyrinato]manganese(III) Chloride (4) and [5,10,15-Tris(ethoxycarbonyl)porphyrinato]-manganese(III) Chloride (5).

Porphyrins **4** and **5** were also isolated during the chromatographic purification of the previous reaction. Porphyrin **4**: mp >300 °C; UV-vis spectroscopy λ_{max} = 460.5 nm, ε = 7.8 x 10^4 L/cm-mole; FAB MS m/z = 623 $[C_{30}H_{24}MnN_4O_8]^+$. Porphyrin **5**: mp >300 °C; UV-vis spectroscopy λ_{max} = 454.5 nm, ε = 1.14 x 10^5 L/cm-mole; FAB MS m/z = 579 $[C_{29}H_{24}MnN_4O_6]^+$.

EXAMPLE 2

II. [5,10,15,20-Tetrakis(ethoxycarbonyl)porphyrinato]zinc(II) (6).



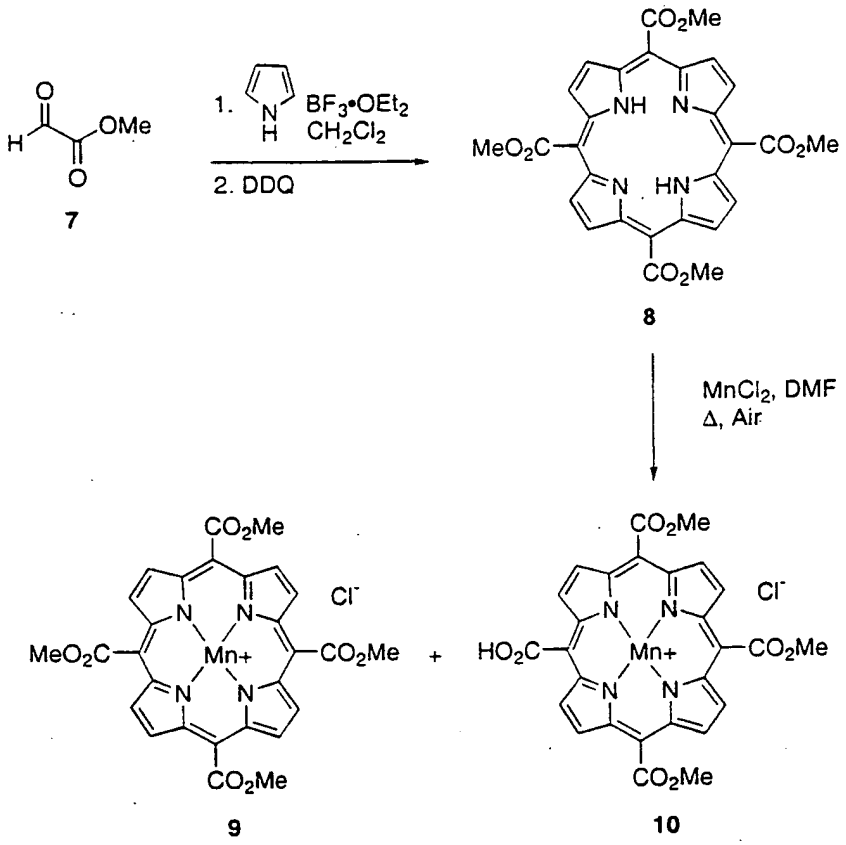
1. [5,10,15,20-Tetrakis(ethoxycarbonyl)porphyrinato]zinc(II) (6).

A solution of **2** (110 mg, 0.18 mmol) and $Zn(OAc)_2$ (403 mg, 18 mmol) in DMF (25 mL) was heated at 145-150 °C for 2 h. The reaction mixture was allowed to cool to room temperature then the DMF was removed by concentration *in vacuo*. The resulting crude solid mixture was adsorbed onto silica gel (3 g) then purified by column chromatography (gradient elution 0→1% $MeOH/CH_2Cl_2$) to provide **6** as a violet solid in 82% yield: mp >300 °C, UV-vis spectroscopy λ_{max} = 412.5 nm, $\epsilon = 2.8 \times 10^5$ L/cm-mole; FAB MS m/z = 660 $[C_{32}H_{28}N_4O_8Zn]^+$; ^1H

NMR (300 MHz, DMSO-d_6) δ 1.79 (t, 12 H), 5.09 (q, 8 H), 9.56 (s, 8 H); ^{13}C NMR (75 MHz, DMSO-d_6) δ 14.50, 63.16, 112.57, 132.03, 147.55, 170.67.

EXAMPLE 3

III. [5,10,15,20-Tetrakis(methoxycarbonyl)porphyrinato]manganese(III) Chloride (9) and [5-Carboxy-10,15,20-tris(methoxycarbonyl)-porphyrinato]manganese(III) Chloride (10).



1. 5,10,15,20-Tetrakis(methoxycarbonyl)porphyrin (8).

In a foil covered, 22 L three-neck round-bottomed flask equipped with a mechanical stirrer and a N_2 inlet was added sequentially, freshly distilled methyl glyoxylate (7) (Hook, J. M. *Synth. Commun.* **1984**, *14*, 83-87) (16.5 g, 187

mmol), CH_2Cl_2 (19 L), and pyrrole (13.0 mL, 194 mmol). The reaction mixture was stirred for 5-10 min then $BF_3 \bullet OEt_2$ (2.30 mL, 18.7 mmol) was added dropwise. After a stirring period of 1.25 h at room temperature, DDQ (31.9 g, 140.4 mmol) was added. The reaction mixture was stirred for an additional 2.25 h at room temperature, then clay (Clarion 550, 25 g) was added and the suspension was stirred for 2.5 h. Filtration of the reaction mixture through Celite provided, after evaporation of solvents, a crude solid mixture which was adsorbed onto silica gel (15 g). Repeated chromatographic purification (5 batches; CH_2Cl_2 as eluent) on silica gel provided porphyrin **8** (1.55 g, 6.1%) as a solid: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.42 (s, 2 H), 4.60 (s, 12 H), 9.48 (2, 4 H); UV-vis λ_{max} = 404.5 nm.

2. [5,10,15,20-Tetrakis(methoxycarbonyl)porphyrinato]manganese(III) Chloride (9).

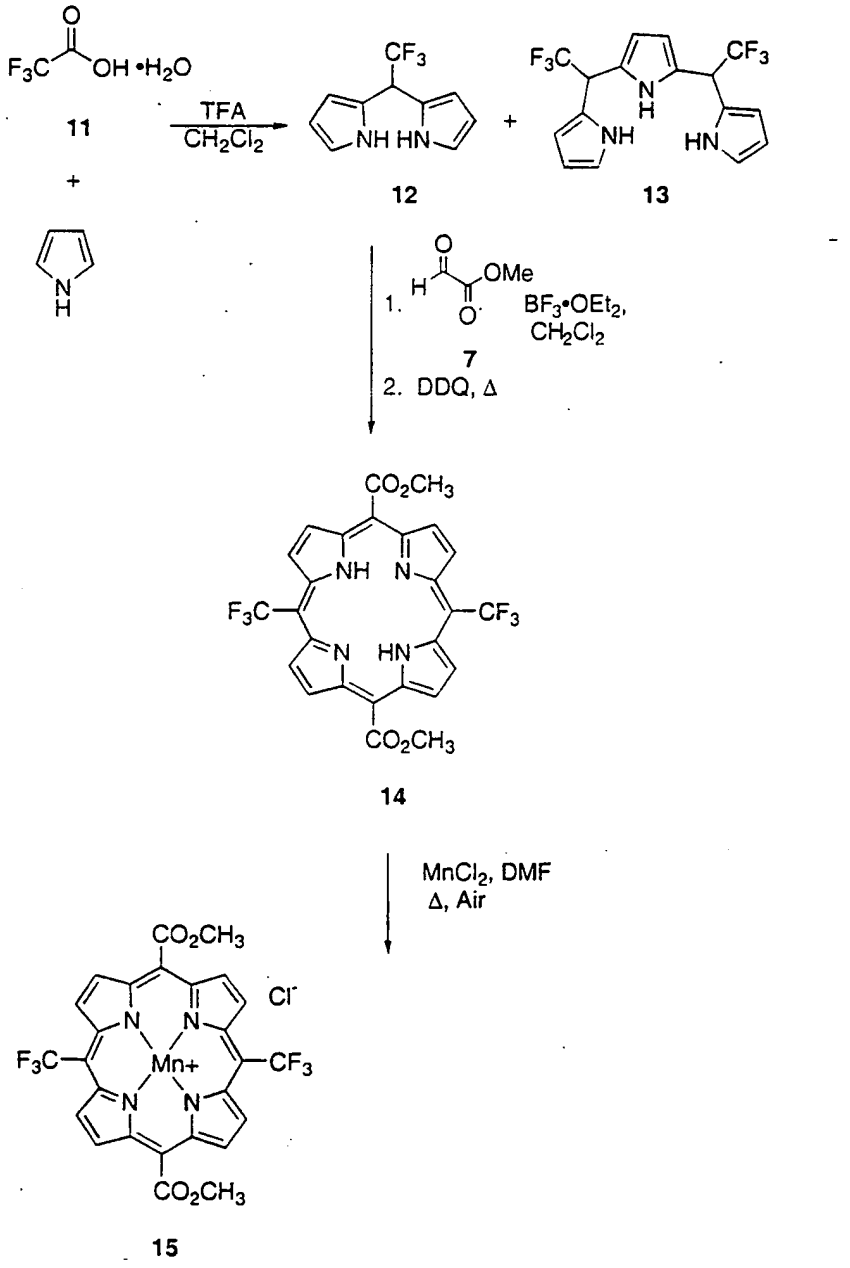
A solution of **8** (1.11 g, 2.0 mmol) and $MnCl_2$ (1.3 g, 10.3 mmol) in DMF (100 mL) was heated at 145 °C for 1-1.5 h then exposed to a stream of air. The reaction mixture was heated for an additional 2-3 h. The reaction mixture was allowed to cool to room temperature overnight under a stream of air. Evaporation of the DMF provided a solid mixture which was adsorbed onto silica gel (3.5 g). Purification by column chromatography (2 batches; gradient elution 0→10% MeOH/CH_2Cl_2) provided porphyrin **9** (760 mg; 59%): mp >300 °C; UV-vis λ_{max} = 455.5 nm, $\epsilon = 8.8 \times 10^4$ L/cm-mole; FAB MS m/z = 595 $[C_{28}H_{20}MnN_4O_8]^+$.

3. [5-Carboxy-10,15,20-tris(methoxycarbonyl)porphyrinato]-manganese(III) Chloride (10).

Porphyrin **10** was also isolated by chromatography from the metalation process above: mp >300 °C; UV-vis λ_{max} = 459.5 nm, $\epsilon = 8.5 \times 10^4$ L/cm-mole; FAB MS m/z = 581 $[C_{27}H_{18}MnN_4O_8]^+$.

EXAMPLE 4

IV. [5,15-Bis(methoxycarbonyl)-10,20-bis(trifluoromethyl)porphyrinato]-manganese(III) Chloride (15) and [5,15-Bis(trifluoromethyl)-10-carboxy-20-(methoxycarbonyl)porphyrinato]manganese(III) Chloride (16).



1. ***meso*-(Trifluoromethyl)dipyrromethane (12)** (Nishino, N.; Wagner, R. W.; Lindsey, J. S.. *J. Org. Chem.* **1996**, *61*, 7534-7544.)

In a 250 mL round-bottomed flask, equipped with a magnetic stirrer and N_2 inlet was placed trifluoracetaldehyde hydrate (**11**, 6.7 g, 58 mmol), pyrrole (8.0 mL, 116 mmol), and CH_2Cl_2 (200 mL). Trifluorocetic acid (4.5 mL, 58 eq) was then added and the reaction mixture was stirred overnight at room temperature. The reaction mixture was transferred to a separatory funnel, then washed consecutively with H_2O (75 mL) and saturated aqueous $NaHCO_3$ (60 mL). The organic layer was dried (Na_2SO_4), filtered, and the solvent removed *in vacuo*. Column chromatography of the residue provided **12** (2.07 g; 17%) and **13**. Dipyrromethane **12**: ^{1}H NMR (300 MHz, $CDCl_3$) δ 4.85 (q, 1 H), 6.19 (m, 4 H), 6.77 (m, 2 H), 8.09 (broad s, 2 H). Tripyrrane **13**: ^{1}H NMR (300 MHz, $CDCl_3$) δ 4.73 (m, 2 H), 6.34 (m, 6 H), 6.75 (m, 2 H), 7.95 (broad s, 1 H), 8.09 (broad s, 2 H); DI MS m/z = 361 $[C_{16}H_{12}N_3F_6 + H]^+$.

2. 5,15-Bis(methoxycarbonyl)-10,20-bis(trifluoromethyl)porphyrin (14).

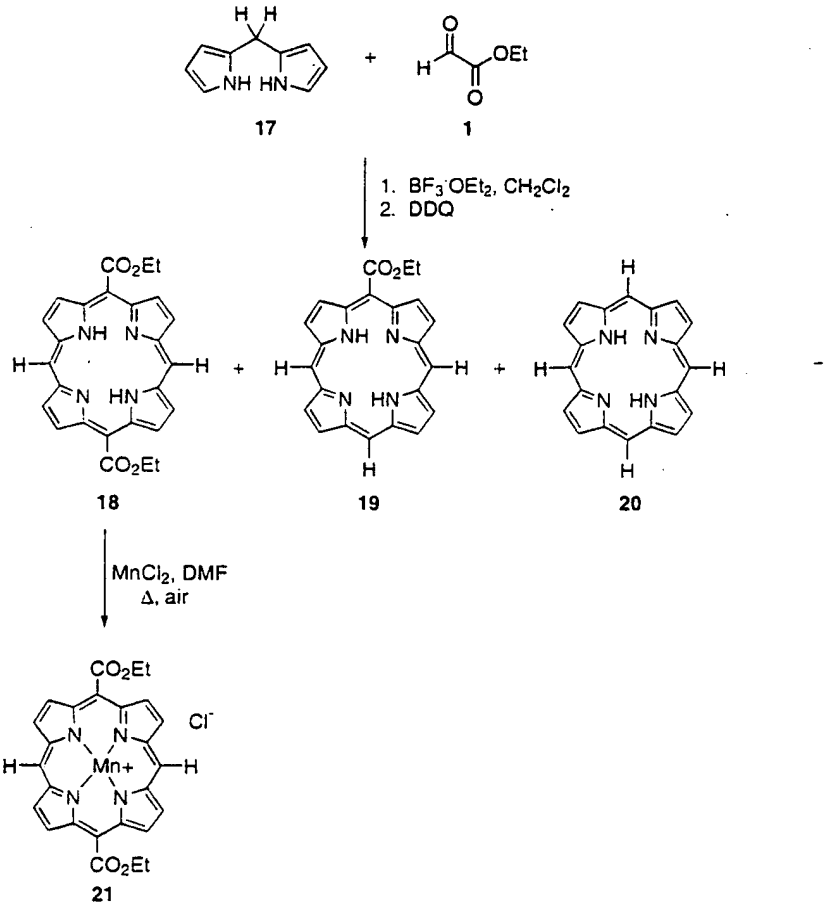
In a foil covered, 500 mL three-neck round-bottomed flask equipped with a magnetic stirrer and a N_2 inlet was added consecutively, freshly distilled methyl glyoxylate (Hook, J. M. *Synth. Commun.* **1984**, *14*, 83-87) (244 mg, 2.77 mmol), dipyrromethane **12** (590 mg, 2.75 mmol) and CH_2Cl_2 (280 mL). The reaction mixture was stirred for 5-10 min then BF_3•OEt_2 (112 mL, 0.9 mmol) was added. After a stirring period of 2 h at room temperature, DDQ (945 mg, 4.1 mmol) was added. The reaction mixture was stirred for an additional 2 h at room temperature, then the solvent was removed *in vacuo*. The residue was absorbed onto silica gel (3.8 g) then purified by column chromatography (CH_2Cl_2 as eluent) to provide porphyrin **14** (390 mg; 30%): ^{1}H NMR (300 MHz, $CDCl_3$) δ -2.96 (s, 2 H), 4.60 (s, 6 H), 9.44 (d, 4 H), 9.73 (m, 4 H).

3. [5,15-Bis(methoxycarbonyl)-10,20-bis(trifluoromethyl)porphyrinato]-manganese(III) Chloride (15).

A solution of **14** (115 mg, 0.20 mmol) and $MnCl_2$ (130 mg, 1.0 mmol) in DMF (30 mL) was heated at 145 °C overnight. The reaction mixture was cooled to room temperature while exposed to a stream of air. Evaporation of DMF provided a solid mixture which was adsorbed onto 3 g silica gel. Purification by column chromatography (gradient elution with 3-10% $MeOH/CH_2Cl_2$) provided porphyrin **15** (117 mg): mp >300 °C; UV-vis λ_{max} = 450 nm, $\epsilon = 9.90 \times 10^4$ L/cm-mol; FAB MS m/z = 615 $[C_{26}H_{14}F_6MnN_4O_4]^+$.

EXAMPLE 5

V. [5,15-Bis(ethoxycarbonyl)porphyrinato]manganese(III) Chloride (21).



1. 5,15-Bis(ethoxycarbonyl)porphyrin (18).

In a foil covered 250 mL round-bottom flask equipped with a magnetic stir bar and a N_2 inlet was added consecutively ethyl glyoxylate (50% in toluene; 310 mg, 1.5 mol), CH_2Cl_2 (150 mL) and dipyrromethane **17** (Chong, *et al., Aust. J. Chem.* **1969**, *22*, 229.) The reaction mixutre was stirred for 5-10 min, then $BF_3{\bullet}OEt_2$ (37 mL, 0.3 mmol) was added. After a 30 min stirring period, DDQ (258 mg, 1.14 mmol) was added and the solution was stirred overnight. The crude material was adsorbed onto silica gel (3 g) and was purified by column chromatography (gradient elution with 50→75% CH_2Cl_2/hexane) to provide

porphyrin **18** (22 mg; 7%): ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.2 (s, 2 H), 1.9 (m, 6 H), 5.15 (m, 4 H), 9.5 (m, 4 H), 9.7 (m, 4 H), 10.4 (s, 2 H).

2. 5-(Ethoxycarbonyl)porphyrin (19) and Porphyrin (20).

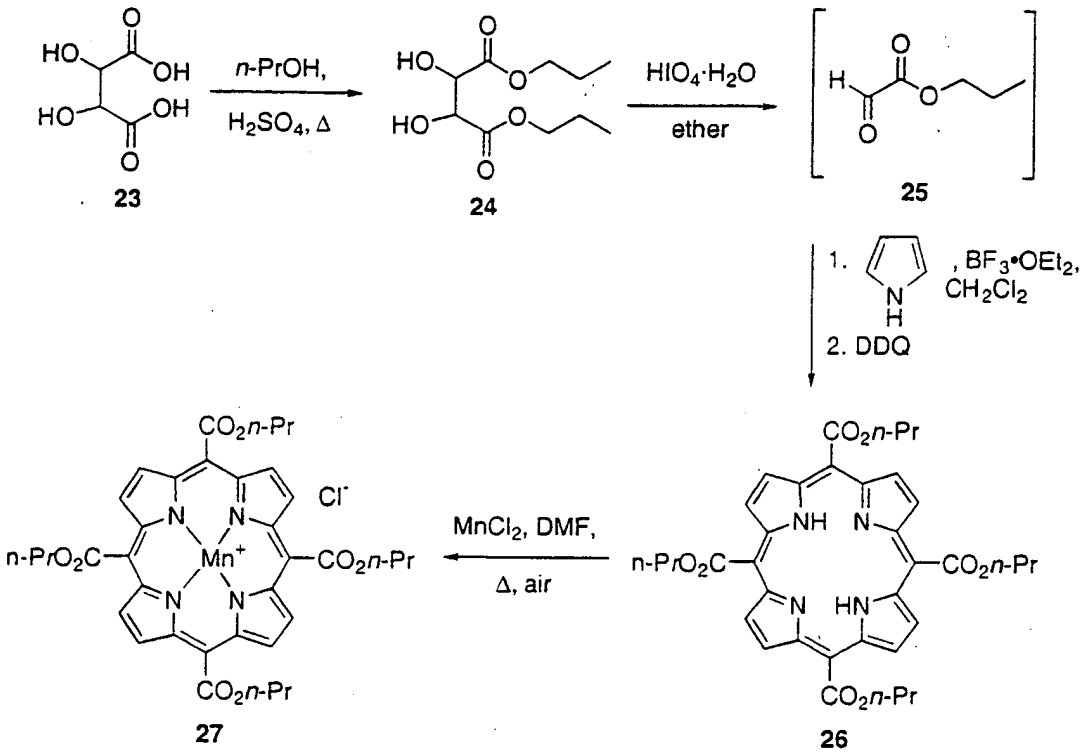
Porphyrins **19** (10 mg; 2%) and **20** (3 mg; 0.5%), were isolated during chormatographic purification of the previous reaction mixture. For porphyrin **19**: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.74 (s, 2 H), 1.87 (t, J = 6.9 Hz), 5.16-5.13 (m, 2 H), 9.43 (s, 4 H), 9.49-9.47 (m, 2 H), 9.65 (m, 2 H), 10.26 (s, 1 H), 10.31 (s, 2 H); FAB MS m/z = 382 $[C_{23}H_{18}N_4O_2]^+$.

3. [5,15-Bis(ethoxycarbonyl)porphyrinato]manganese(III) Chloride (21).

A solution of porphyrin **18** (22 mg, 0.058 mmol), $MnCl_2$ (55 mg, 0.44 mmol) and anhydrous DMF (8 mL) was heated at 125 °C in a round-bottom flask fitted with a reflux condenser. After 1 h, the reaction mixture was exposed to a stream of air and the solution was stirred overnight. The reaction mixture was allowed to cool to room temperature, then the solvent was evaporated *in vacuo*. The crude solid material was absorbed onto silica gel (1.2 g) and purified by column chromatography (gradient elution 0→7% MeOH/CH_2Cl_2) to provide **21** (6 mg; 19%) as a dark solid: mp >300 °C; UV-vis λ_{max} = 452.5 nm, ε = 2.48 x 10^4 L/cm-mol; FAB MS m/z = 507 $[C_{26}H_{20}MnN_4O_4]^+$.

EXAMPLE 6

VI. [5,10,15,20-Tetrakis(*n*-propoxycarbonyl)porphyrinato]manganese(III) Chloride (27).



23 24 25 26 27

1. Di-*n*-propyl *d*-tartrate (24).

d-Tartaric acid (2.56 g, 17.0 mmol), *n*-propanol (30 mL) and concentrated H_2SO_4 (6 mL) were heated under reflux for 3 d. The solution was cooled to room temperature then partitioned between H_2O and CH_2Cl_2. The organic layer was washed consecutively with aqueous saturated $NaHCO_3$, H_2O, and brine, then dried (Na_2SO_4), filtered, and the solvent removed *in vacuo*. The resulting crude ester **24** (2.6 g, 65%) was used without further purification: ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.0 (t, 6 H), 1.75 (m 2 H), 3.2 (two singlets, 2 H), 4.25 (m, 4 H), 4.55 (m 2 H); DI MS m/z = 235 $[C_{10}H_{18}O_6+H]^+$.

2. *n*-Propyl Glyoxylate (25)

A solution of di-*n*-propyl *d*-tartrate (**24**, 0.73 g, 3.1 mmol) in anhydrous ether (10 mL) was magnetically stirred at 0 °C under dry N_2, as periodic acid dihydrate (0.711 g, 3.12 mmol) was added. The resulting solution was stirred for 1 h. Anhydrous Na_2SO_4 was added and the resulting milky solution was filtered, and the solvent was evaporated *in vacuo* to provide **25** as an oil (0.670 g; 93%): CI MS (methane) m/z = 117 $[C_5H_8O_3+H]^+$.

3. 5,10,15,20-Tetrakis(*n*-propoxycarbonyl)porphyrin (26).

To a foil covered, 2 L three-neck round-bottom flask equipped with a magnetic stirrer and a N_2 inlet was added consecutively **25** (1.31 g, 11.3 mmol), CH_2Cl_2 (1.1 L) and pyrrole (0.78 ml, 11.2 mmol). The reaction mixture was stirred for 5 min, then $BF_3 \bullet OEt_2$ (416 mL, 3.3 mmol) was added. After a stirring period of 3.5 h, DDQ (1.92 g, 8.5 mmol) was added to the reaction mixture and stirring continued for an additional 2 h at room temperature. Clay (Clarion 550 clay, 6 g) was added and the reaction mixture was allowed to stir overnight. Filtration of the heterogeneous mixture through Celite followed by removal of the solvent *in vacuo* provided a solid mixture which was adsorbed onto silica gel (2.5 g). Chromatographic purification (gradient elution 66-100% CH_2Cl_2/hexanes) provided **27** (130 mg; 7%) as a solid: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.31 (s, 2 H), 1.29 (t, 12 H), 2.18 (m, 8 H), 5.02 (7, 8 H), 9.52 (s, 8 H).

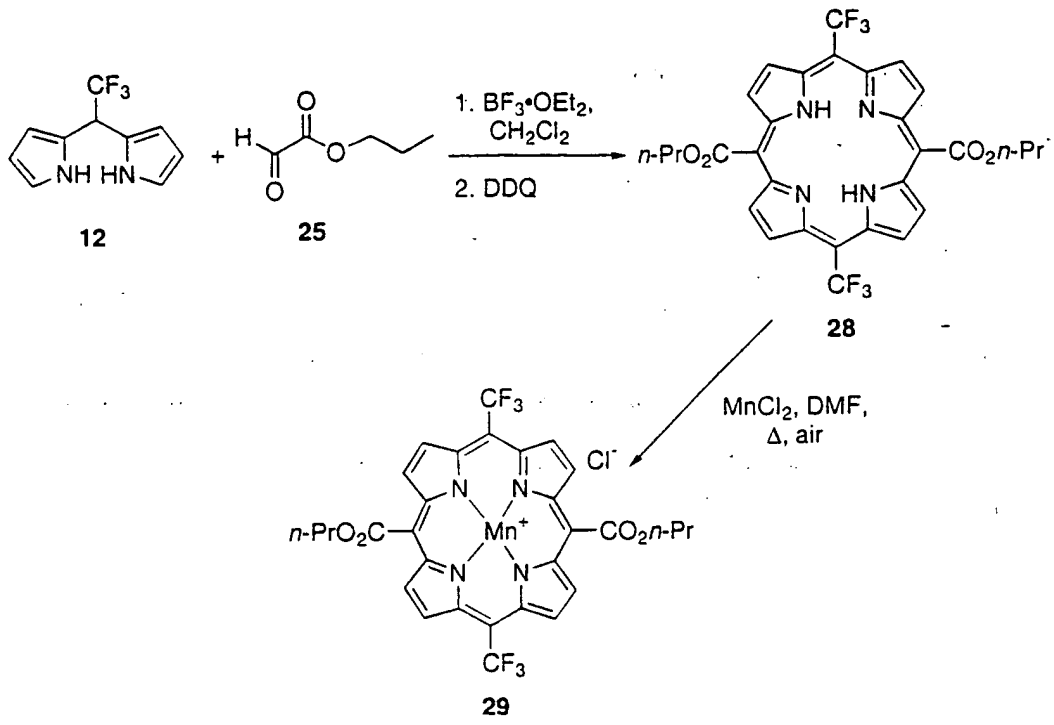
4. [5,10,15,20-Tetrakis(*n*-propoxycarbonyl)porphyrinato]manganese(III) Chloride (27).

A solution of porphyrin **26** (170 mg, 0.260 mmol) and $MnCl_2$ (167 mg, 1.32 mmol) in DMF was heated at 145 °C for 2 h then exposed to a stream of air. The reaction mixture was heated for another 2-3 h then cooled to room temperature overnight under a stream of air. Evaporation of DMF *in vacuo*

provided a solid mixture which was adsorbed onto silica gel (2.6 g). Purification by column chromatography (elution with 2.5% MeOH/CH_2Cl_2) provided **27** (170 mg; 88%) as a dark solid: mp >300 °C; UV-vis λ_{max} = 456 nm, ϵ = 1.35 x 10^5 L/cm-mol; FAB MS m/z = 707 $[C_{36}H_{36}MnN_4O_8]^+$.

EXAMPLE 7

VII. [5,15-Bis(*n*-propoxycarbonyl)-10,20-bis(trifluoromethyl)porphyrinato]-manganese(III) Chloride (29).



1. 5,15-Bis(*n*-propoxycarbonyl)-10,20-bis(trifluoromethyl)porphyrin (28).

In a foil covered, 250 mL three-neck round-bottomed flask equipped with a magnetic stirrer and a N_2 inlet was added consecutively *n*-propyl glyoxylate (**25**; 154 mg, 1.3 mmol), dipyrromethane **12** (283 mg; 1.3 mmol) and CH_2Cl_2 (130 mL). The reaction mixture was stirred for 5-10 min then $BF_3 \bullet OEt_2$ (32 μL, 0.26 mmol) was added. After 1.5 h at room temperature, DDQ (225 mg, 1.0 mmol) was added. The reaction mixture was stirred for an additional 2 h at room temperature, then treated with clay (Clarion 550, 500 mg). Filtration of the reaction mixture through Celite, followed by evaporation of solvents provided a residue which was absorbed onto silica gel (2.5 g). Column chromatography

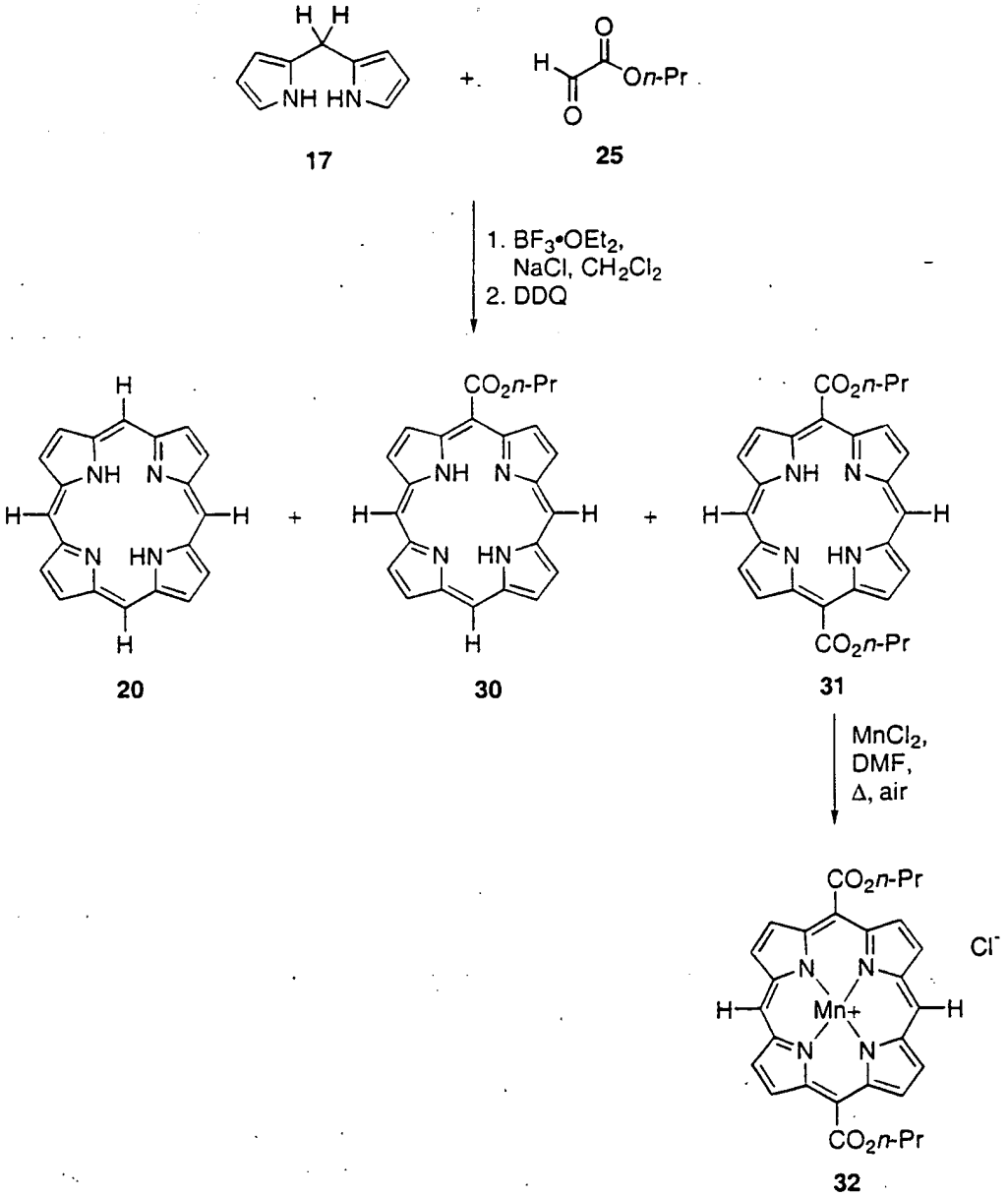
(elution with 50% CH_2Cl_2/hexanes) provided porphyrin **28** (15 mg; 3.7%) as a solid: ^{1}H NMR (300 MHz, $CDCl_3$) δ -2.94 (s, 2 H), 1.3 (m, 6H), 2.2 (m, 4 H), 5.04 (t, 4 H), 9.46 (d, 4 H), 9.74 (m, 4 H).

2. [5,15-Bis(*n*-propoxycarbonyl)-10,20-bis(trifluoromethyl)porphyrinato]-manganese(III) Chloride (29).

A solution of **28** (15 mg, 0.02 mmol) and $MnCl_2$ (28 mg, 0.22 mmol) in DMF (10 mL) was heated at 145 °C while exposed to a stream of air. After analysis by UV-vis indicated that the reaction was complete, the reaction mixture was cooled to room temperature. Evaporation of DMF provided a solid mixture which was absorbed onto silica gel (1.5 g). Purification by column chromatography (gradient elution with 0→2.5% MeOH/CH_2Cl_2) provided porphyrin **28** (15 mg; 87%): UV-vis λ_{max} = 450.5 nm, $\epsilon = 1.10 \times 10^5$ L/cm-mol; FAB MS m/z = 671 $[C_{30}H_{22}F_6MnN_4O_4]^+$.

EXAMPLE 8

VIII. [5,15-Bis(*n*-propoxycarbonyl)porphyrinato]manganese(III) Chloride (32) and [5-(*n*-propoxycarbonyl)porphyrinato]manganese(III) Chloride (33).



1. 5-(*n*-Propoxycarbonyl)porphyrin (30) and 5,15-Bis(*n*-propoxycarbonyl)porphyrin (31).

To a foil covered round-bottom flask equipped with a magnetic stirrer and a N_2 inlet was added consecutively dipyrromethane **17** (Chong, *et al., Aust. J. Chem.* **1969**, *22*, 229) (0.63 g, 4.3 mmol), CH_2Cl_2 (430 mL), *n*-propyl glyoxylate (**25**, 0.5 g, 4.3 mmol) and NaCl (23 mg, 0.43 mmol). The reaction mixture was stirred for 5-10 min, then $BF_3 \bullet OEt_2$ (160 mL, 1.3 mmol) was added. After a 65 min stirring period, DDQ (732 mg, 3.23 mmol) was added and the reaction mixture was stirred overnight. Removal of solvents *in vacuo* provided a crude product which was adsorbed onto silica gel (2 g). Column chromatographic purification (gradient elution with 50%→80% CH_2Cl_2/hexane) afforded porphyrin **20**, porphyrin **30** (10 mg, 1.2%) and porphyrin **31** (30 mg, 2.9%): For porphyrin **30**: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.62 (s, 2 H), 1.31 (t, 3 H), 2.24 (m, 2 H), 5.04 (t, 2 H), 9.48 (d, 2H), 9.51 (m, 4 H), 9.70 (d, 2 H), 10.33 (s, 1 H), 10.36 (s, 2 H). For porphyrin **31**: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.33 (s, 2 H), 1.31 (t, 6 H), 2.24 (m, 4 H), 5.03 (t, 4 H), 9.47 (d, 4 H), 9.69 (d, 4 H), 10.38 (2 H).

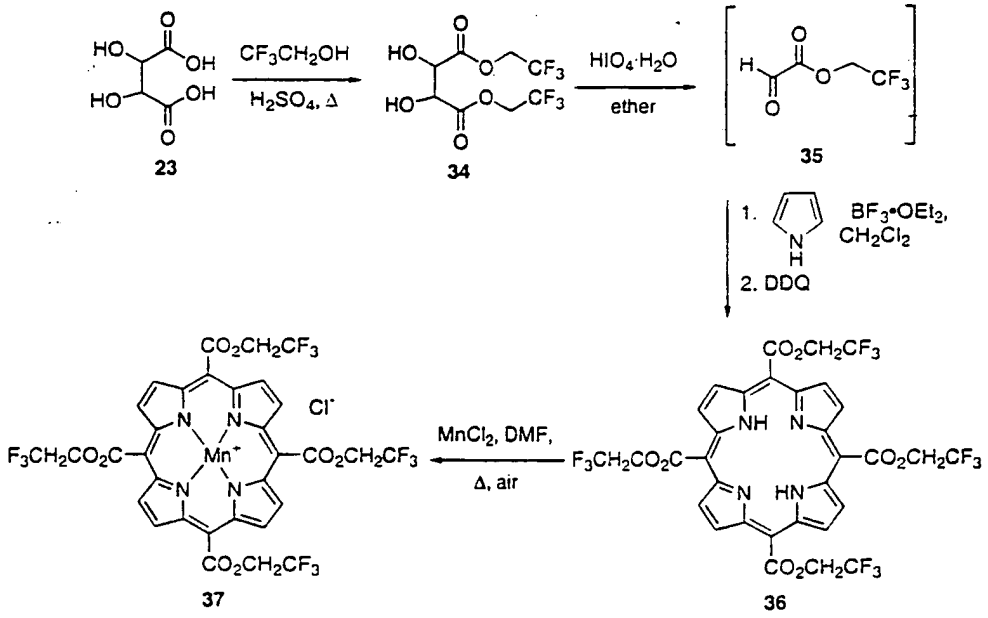
2. [5,15-Bis(*n*-propoxycarbonyl)porphyrinato]manganese(III) Chloride (32).

To a magnetically stirred solution of porphyrin **31** (30 mg, 0.062 mmol) in anhydrous DMF (25 mL) was added $MnCl_2$ (39 mg, 0.27 mmol). The reaction flask was fitted with a reflux condenser and the solution was heated to 145 °C for 2 h then exposed to a stream of air. The reaction mixture was heated for an additional 3-4 h then the solution was allowed to cool to room temperature for 48 h under a stream of air. Evaporation of DMF *in vacuo* provided a crude product that was adsorbed onto silica gel (2 g). Purification by column chromatography (gradient elution with 5→7.5% MeOH/CH_2Cl_2) provided **30**

(35 mg; 98%) as a dark solid: mp >300 °C; UV-vis λ_{max} = 452.5 nm, ε = 6.10 x 10^4 L/cm-mole; FAB MS m/z = 535 [$C_{28}H_{24}MnN_4O_4]^+$.

EXAMPLE 9

IX. [5,10,15,20-Tetrakis(2,2,2-trifluoroethoxycarbonyl)porphyrinato]-manganese(III) Chloride (37).



1. Di-(2,2,2-trifluoroethyl) *d*-tartrate (34).

d-Tartaric acid (**23**; 25g, 0.17 mol), trifluoroethanol (200 ml) and H_2SO_4 (59 mL) were magnetically stirred and heated under reflux for 22 h. The solution was cooled to room temperature, partitioned between H_2O and CH_2Cl_2. The H_2O layer was extracted with CH_2Cl_2 (2 x 150 mL). The combined organic layers were washed with saturated aqueous $NaHCO_3$, and brine, then filtered through a bed of Celite and evaporated *in vacuo*. The residue was dissolved in ethyl ether and filtered through a plug of silica. The silica was rinsed with ca.

600 mL. The ether was removed *in vacuo* and the residue was recrystallized from ether/hexane to yield **34** (17.3; 33%) as white crystals: mp 77-79 °C; ^{1}H NMR (300 MHz, $CDCl_3$) δ 3.13 (two singlets, 2H), 4.6 (m, 6 H).

2. 2,2,2-Trifluoroethyl glyoxylate (35).

A solution of **34** (1 g, 3.2 mmol) in anhydrous ether (25 mL) was magnetically stirred at 0 °C under dry N_2 as periodic acid dihydrate (0.72 g, 3.2 mmol) was added in portions (3 x 0.24 g) over the course of 1 h. The solution was stirred for an additional 4 h. The reaction solution was decanted, dried (Na_2SO_4), filtered and the solvent removed *in vacuo*. The material was used immediately and without further purification (0.85 g, 86%).

3. 5,10,15,20-Tetrakis(2,2,2-trifluoroethoxycarbonyl)porphyrin (36).

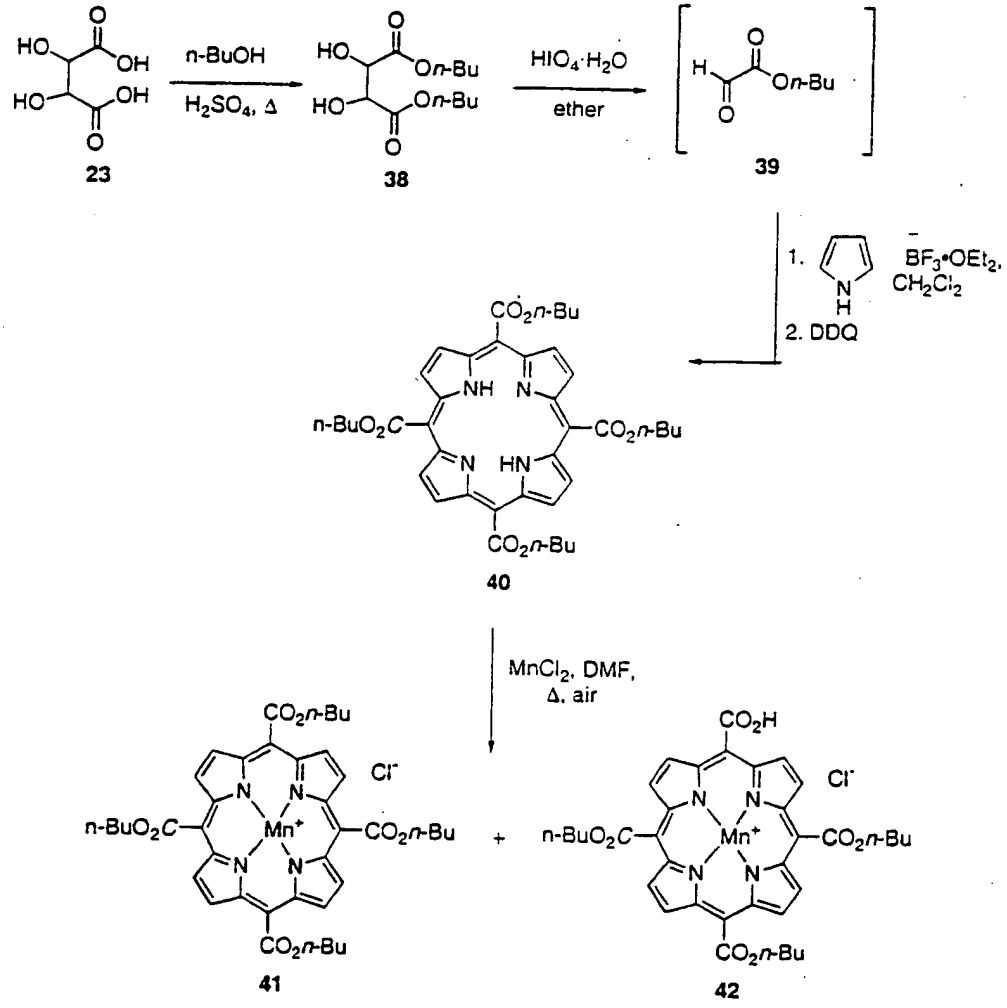
To a foil covered round-bottomed flask equipped with a magnetic stirrer and N_2 inlet was added sequentially 2,2,2-trifluoroethyl glyoxylate **33** (0.85 g, 5.4 mmol), CH_2Cl_2 (550 mL) and pyrrole (0.38 mL, 5.4 mmol). The reaction mixture was stirred for 5 min then BF_3•OEt_2 (0.20 mL, 1.62 mmol) was added. The reaction mixture was allowed to stir. After a 1.5 h stirring period, DDQ (0.927g, 4mmol) was added and the reaction mixture was allowed to stir an additional 30 min. Clay (Clarion 550, 4.17 g) was added and the resulting suspension was stirred for 1 h, then the entire solution was filtered through a bed of Celite. The filtrate was evaporated and adsorbed onto silica gel (2.5 g). Chromatography of the residue on silica (elution with 50-66% CH_2Cl_2/hexanes) provided **36** (37 mg; 3.4%) as a solid: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.6 (s, 2 H), 5.5 (q, 8 H), 9.5 (s, 8 H).

4. [5,10,15,20-Tetrakis(2,2,2-trifluoroethoxycarbonyl)porphyrinato]-manganese(III) Chloride (37).

To a round-bottom flask equipped with a magnetic stirrer and a condenser was added porphyrin **36** (36 mg, 0.043 mmol), $MnCl_2$ (27 mg, 0.21 mmol) and DMF (10 ml). The reaction mixture was heated to 140 °C for 2 h, then exposed to a stream of air. After 5 h, an additional amount of $MnCl_2$ (27 mg, 0.21 mmol) was added and the reaction mixture was stirred for 3 d at 100 °C. The solvent was removed *in vacuo* and the resudue was adsorbed onto silica gel (1 g). Chromatography on silica gel (gradient elution 0→3% MeOH/CH_2Cl_2) provided porphyrin **37** (23 mg; 58%) as a dark solid: FAB MS m/z = 867 $[C_{32}H_{16}F_{12}MnN_4O_8]^+$; UV-vis λ_{max} = 453.5 nm, $\epsilon = 7.30 \times 10^4$ L/cm-mol.

EXAMPLE 10

X. [5,10,15,20-Tetrakis(*n*-butoxycarbonyl)porphyrinato]manganese(III) Chloride (41) and [5-Carboxy-10,15,20-tris(*n*-butoxycarbonyl)-porphyrinato]manganese(III) Chloride (42).



1. Di-*n*-butyl *d*-tartrate (38)

d-Tartaric acid (25 g, 167 mmol), *n*-butanol (200 mL) and concentrated H_2SO_4 (59 mL) were heated under reflux overnight. The solution was cooled to room temperature then partitioned between H_2O and CH_2Cl_2. The organic layer was washed with aqueous saturated $NaHCO_3$ then filtered through Celite. The organic solution was then washed with brine, dried ($MgSO_4$), filtered, and the solvent removed *in vacuo*. The resulting oil was purified by Kugelrohr distillation (bp = 100-105 °C, 0.06 mm Hg) to yield **38** as a clear light yellow oil: ^1H NMR (300 MHz, $CDCl_3$) δ 0.9 (t, 6 H), 1.4 (m, 4 H), 1.65 (m, 4 H), 3.2 (m, 2 H), 4.35 (m, 4 H), 4.5 (m, 2 H).

2. *n*-Butyl Glyoxylate (39)

A solution of tartrate **38** (5 g, 19 mmol) in anhydrous ether (150 mL) was magnetically stirred at 0 °C and under dry N_2, as periodic acid dihydrate (4.35 g, 19 mmol) was added over 1 h in portions (3 x 1.45 g). The resulting solution was stirred for 4 h, decanted from the solid precipitate, dried (Na_2SO_4), filtered, and the solvent was removed *in vacuo* to provide **39** (4.72 g; 96%) as an oil. The crude mixture was used immediately without futher purification.

3. 5,10,15,20-Tetrakis(*n*-butoxycarbonyl)porphyrin (40).

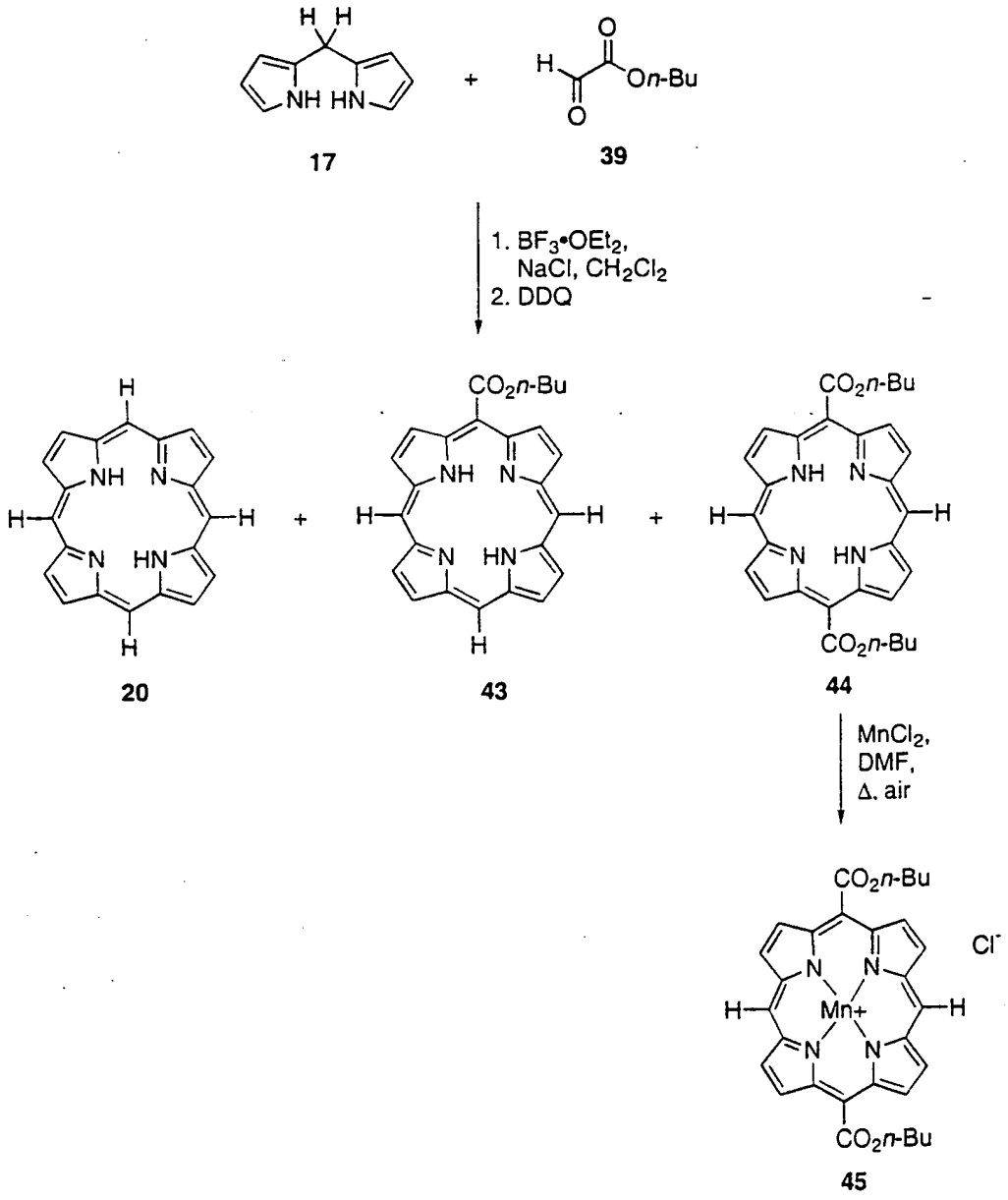
To a foil covered round-bottomed flask equipped with a magnetic stirrer and a N_2 inlet was added *n*-butyl glyoxylate (**39**, 2.5 g, 19 mmol), CH_2Cl_2 (1.9 L), NaCl (0.11 g, 1.9 mmol) and pyrrole (1.33 ml, 19 mmol). The reaction mixture was stirred for 5 min then $BF_3 \bullet OEt_2$ (0.71 ml, 5.7 mmol) was added. After a stirring period of 1 h, DDQ (3.27 g, 14.3 mmol) was added and the reaction mixture was stirred overnight. Clarion 550 clay (15 g) was added and the resulting suspension was allowed to stir for 2-3 h, then filtered through a pad of Celite. The solvent was removed *in vacuo* and the residue was adsorbed onto silica gel (2 g). Purification by column chromatography (gradient elution with 50→100% CH_2Cl_2/hexanes) afforded porphyrin **40** (0.36 g; 11%) as a dark violet solid: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.32 (s, 2 H), 1.12 (t, 12 H), 1.70 (m, 8 H), 2.14 (q, 8 H), 5.06 (t, 8 H), 9.51, (s, 8 H).

4. [5,10,15,20-Tetrakis(*n*-butoxycarbonyl)porphyrinato]manganese(III) Chloride (41) and [5-Carboxy-10,15,20-tris(*n*-butoxycarbonyl)-porphyrinato]manganese(III) Chloride (42).

A solution of porphyrin **40** (355 mg, 0.50 mmol) and $MnCl_2$ (318 mg, 2.53 mmol) in anhydrous DMF (50 mL) was magnetically stirred and heated to 145 °C for 1 h then exposed to a stream of air for 2 h. The reaction mixture was cooled to room temperature overnight under a stream of air. Evaporation of DMF provided a crude solid mixture which was adsorbed onto silica gel (2 g). Purification by column chromatography (gradient elution with 0-5% MeOH/CH_2Cl_2) provided porphyrins **41** (170 mg) and **42** (10 mg) as solids. For porphyrin **41**: mp 200-210 °C; UV-vis λ_{max} = 456.0 nm, $\epsilon = 1.4 \times 10^5$ L/cm-mol; FAB MS m/z = 763 $[C_{40}H_{44}MnN_4O_8]^+$. For porphyrin **42**: mp 200-205 °C; UV-vis λ_{max} = 459.5 nm, ϵ = 7.2 x 104 L/cm-mol; FAB MS m/z = 707, $[C_{36}H_{36}MnN_4O_8]^+$.

EXAMPLE 11

XI. [5,15-Bis(*n*-butoxycarbonyl)porphyrinato]manganese(III) Chloride (45).



1. 5,15-Bis(*n*-butoxycarbonyl)porphyrin (44).

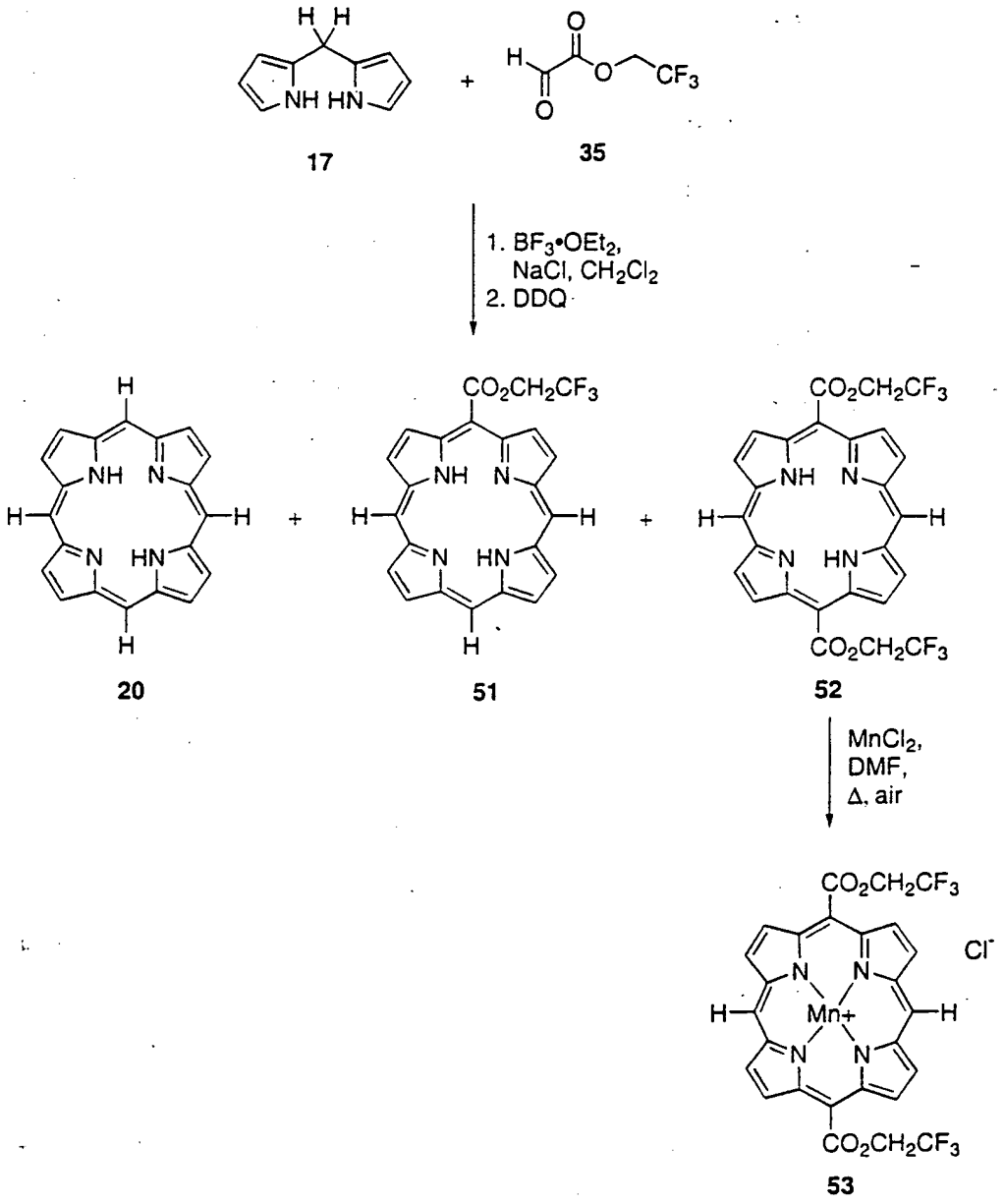
To a foil-covered round-bottom flask equipped with a magnetic stirrer and a N_2 inlet was added sequentially *n*-butyl glyoxylate (**39**, 1.0 g, 7.7 mmol), CH_2Cl_2 (390 mL), NaCl (42 mg, 0.77 mmol) and dipyrromethane (1.12 g, 7.7 mmol). The solution was stirred for 5 min, then $BF_3 \bullet OEt_2$ (283 μL, 2.3 mmol) was added and reaction mixture allowed to stir for 30 min. DDQ (1.3 g, 5.8 mmol) was added, and the reaction mixture was stirred for another 30 min. The solvent was removed *in vacuo* and the residue was adsorbed onto silica gel (2 g). Purification by column chromatography (gradient elution 50→66% CH_2Cl_2/hexane) provided **44** (137 mg; 7%): ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.7 (s, 2 H), 1.16 (t, 6 H), 1.78 (m, 4 H), 2.20 (m, 4 H), 5.07 (t, 4 H), 9.37 (d, 4 H), 9.58 (d, 4 H), 10.19 (s, 2H).

2. [5,15-Bis(*n*-butoxycarbonyl)porphyrinato]manganese(III) Chloride (45).

To a round-bottom flask equipped with a magnetic stirrer was added porphyrin **43** (137 mg, 0.27 mmol), $MnCl_2$ (170 mg, 1.3 mmol) and DMF (110 ml). The reaction mixture was heated to 140 °C. After a stirring period of 2 h, the reaction was exposed to a stream of air overnight at 110 °C. The solvent was removed *in vacuo* and the residue was adsorbed onto silica gel. Purification by column chromatography on silica (elution with 5% MeOH/CH_2Cl_2) provided **45** (65 mg; 43%) as a dark solid: mp > 300 °C; UV-vis λ_{max} = 563 nm, ε = 8.4 x 10^4 L/cm-mole; FAB MS m/z = 563 $[C_{30}H_{28}MnN_4O_4]^+$.

EXAMPLE 12

XIII. [5,15-Bis(trifluoroethoxycarbonyl)porphyrinato]manganese(III) Chloride (53) and [5-(trifluoroethoxycarbonyl)porphyrinato]-manganese (III) Chloride (54).



1. 5-Trifluoroethoxycarbonylporphyrin (51) and 5,15-Bis(trifluoroethoxycarbonyl)porphyrin (52).

To a foil covered round-bottom flask equipped with a magnetic stirrer and a N_2 inlet was added consecutively dipyrromethane **17** (Chong, *et al., Aust. J. Chem.* **1969**, *22*, 229) (1.41 g, 9.6 mmol), CH_2Cl_2 (1.15 L), trifluoroethyl glyoxylate (**35**, 1.6 g, 9.6 mmol) and NaCl (53 mg, 0.96 mmol). The reaction mixture was stirred for 5-10 min, then $BF_3 \bullet OEt_2$ (176 mL, 1.43 mmol) was added. After a 30 min stirring period, DDQ (1.63 g, 7.2 mmol) was added and the reaction mixture was stirred overnight. Removal of solvents *in vacuo* provided a crude product which was adsorbed onto silica gel (2 g). Column chromatographic purification (gradient elution with 50%→80% CH_2Cl_2/hexane) afforded porphyrin **20**, porphyrin **51** (38 mg) and porphyrin **52** (54 mg). For porphyrin **51**: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.58 (s, 2 H), 5.43 (q, 2 H), 9.45 (m, 4 H), 9.51 (d, 2 H), 9.70 (d, 2 H), 10.30 (s, 1 H), 10.35 (s, 2 H). For porphyrin **52**: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.11 (s, 2 H), 5.44 (q, 4 H), 9.50 (d, 4 H), 9.70 (d, 4 H), 10.42 (s, 2 H).

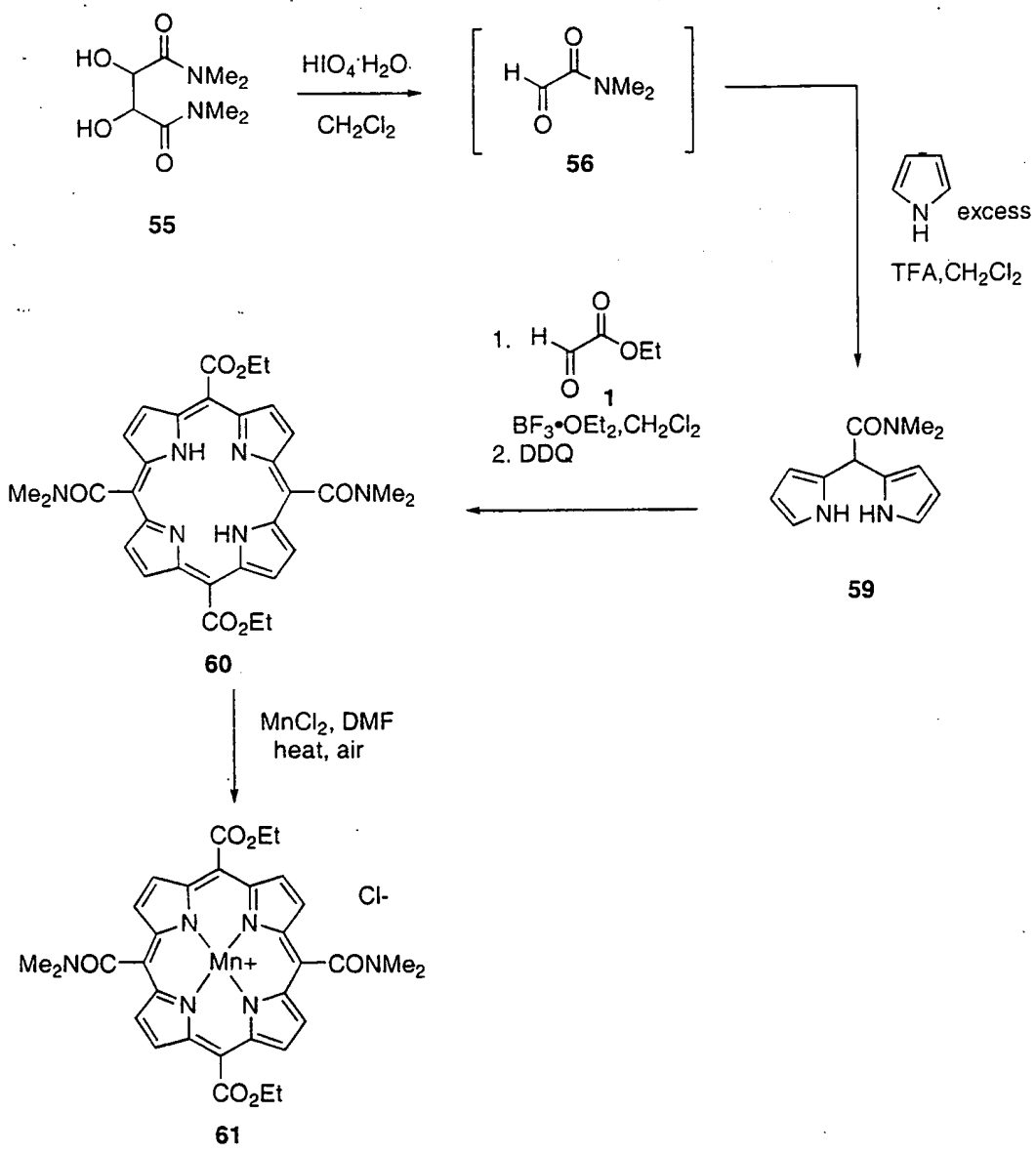
2. [5,15-Bis(trifluoroethoxycarbonyl)porphyrinato]manganese(III) Chloride (53).

To a magnetically stirred solution of porphyrin **31** (54 mg, 0.1 mmol) in anhydrous DMF (60 mL) was added $MnCl_2$ (63 mg, 0.5 mmol). The reaction flask was fitted with a reflux condenser and the solution was heated to 145 °C for 2 h then exposed to a stream of air. Additional $MnCl_2$ (63 mg, 0.5 mmol) was added for completion of the reaction. The reaction mixture was heated for an additional 3-4 h then the solution was allowed to cool to room temperature for 48 h under a stream of air. Evaporation of DMF *in vacuo* provided a crude product that was adsorbed onto silica gel (2 g). Purification by column chromatography (gradient elution with 5→7.5% MeOH/CH_2Cl_2) provided **53**

(45 mg; 72%) as a dark solid: mp >300 °C; UV-vis λ_{max}= 451 nm, $\epsilon = 8 \times 10^4$ L/cm-mole; FAB MS m/z = 615 $[C_{26}H_{14}F_6MnN_4O_4]^+$

EXAMPLE 13

XV. [5,15-Bis(dimethylamido)-10,20-bis(ethoxycarbonyl)porphyrinato]-manganese(III) Chloride (61).



1. *meso*-(*N,N*-Dimethylamido)dipyrromethane (59).

In a round-bottom flask equipped with a magnetic stir bar and N_2 inlet was placed *N,N*-dimethyl glyoxamide (**56**, 2.0 g, 20 mmol), pyrrole (16 mL, 235 mmol) and CH_2Cl_2 (40 mL). Trifluoroacetic acid (0.6 mL, 7.8 mmol) was then added. The resulting red-orange solution was stirred overnight at room temperature, transferred into a separatory funnel, diluted with CH_2Cl_2, then washed with H_2O, saturated aqueous $NaHCO_3$, H_2O and brine. The organic portion was dried (Na_2SO_4) and filtered. The solvent was removed *in vacuo* and the residue was adsorbed onto silica gel (5 g). Purification by chromatography (SiO_2, gradient elution with 40% CH_2Cl_2/hexanes→3% CH_3OH/CH_2Cl_2) afforded dipyrromethane **59** as a light brown solid (1.24 g; 31%): ^{1}H NMR (300 MHz, $CDCl_3$) δ 2.99 (s, 3 H), 3.18 (s, 3 H), 5.41 (s, 1 H), 6.03 (broad s, 2 H), 6.07 (m, 2 H), 6.62 (broad s, 2 H), 9.30 (broad s, 2 H).

2. 5,15-Bis(dimethylamido)-10,20-bis(ethoxycarbonyl)porphyrin (60).

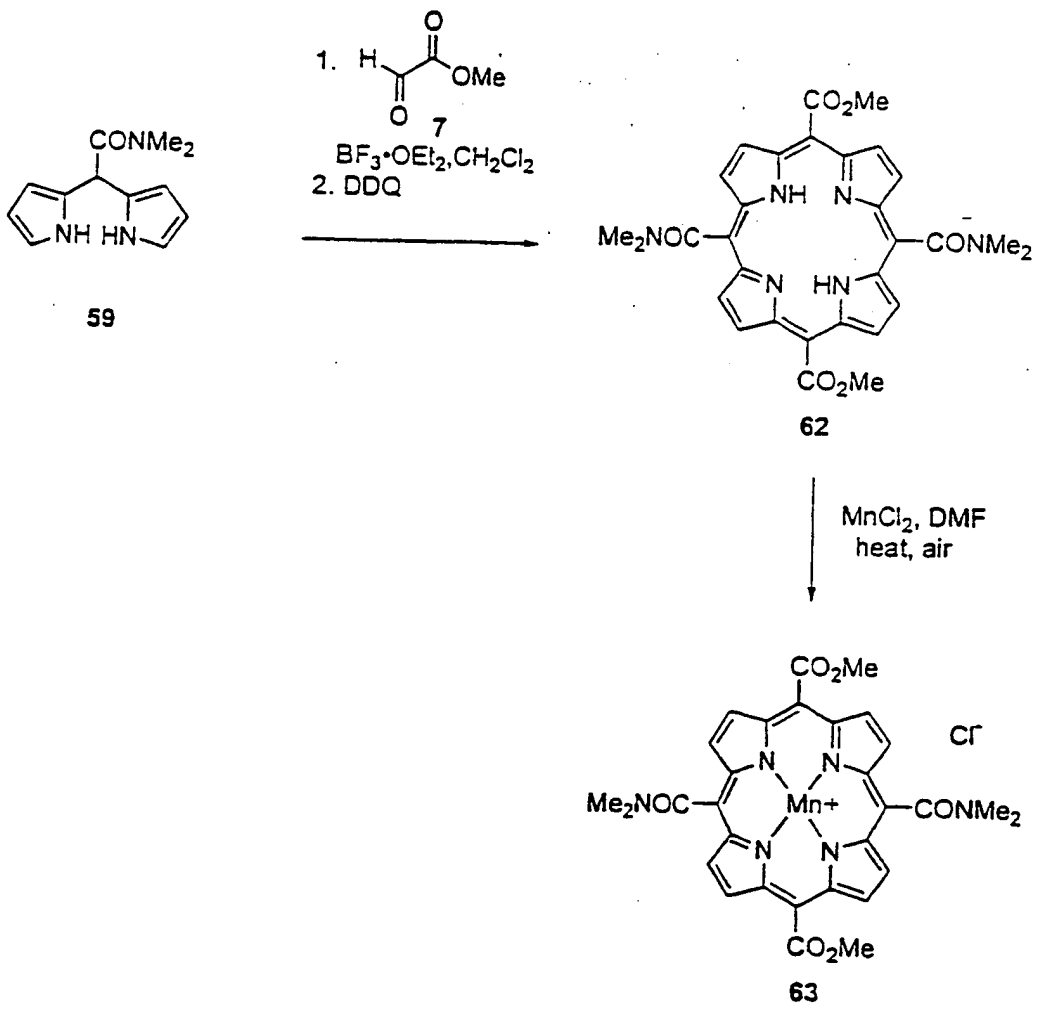
To a foil covered round-bottom flask equipped with a magnetic stirrer and a N_2 inlet was added consecutively dipyrromethane **59** (500 mg, 2.30 mmol), CH_2Cl_2 (230 mL), ethyl glyoxylate (50% in toluene, 470 mg, 2.30 mmol). The reaction mixture was stirred for 5-10 min, then $BF_3 \bullet OEt_2$ (57 μL, 0.46 mmol) was added. After a 45 min stirring period, DDQ (392 mg, 1.73 mmol) was added and the reaction mixture was stirred overnight. The solvent was removed *in vacuo* and the residue was adsorbed onto silica gel (4 g). Repeated purification by column chromatography (gradient elution with 0→3% CH_3OH/CH_2Cl_2) afforded porphyrin **60** (7 mg; 1%) as a dark violet solid and as a mixture of atropisomers: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.2 (s, 2 H), 1.8 (t, 6 H), 2.8 (m, 6 H), 3.8 (s, 6 H), 5.1 (m, 4 H), 9.3 (d, 4 H), 9.5 (d, 4 H); FAB MS m/z = 597 $[C_{32}H_{32}N_6O_6 + H]^+$.

3. [5,15-Bis(dimethylamido)-10,20-bis(ethoxycarbonyl)porphyrinato]-manganese(III) Chloride (61).

A solution of **60** (7.0 mg, 0.012 mmol) and $MnCl_2$ (15 mg, 0.12 mmol) in DMF (6 mL) was heated at 145 °C for 1 h then exposed to a stream of air. The reaction mixture was heated for an additional 1.5 h then allowed to cool to room temperature overnight and under a stream of air. Additional $MnCl_2$ (15 mg, 0.12 mmol) was added to the reaction mixture and heated at 145 °C for 1.5 h then exposed to a stream of air for 20 min while hot. The reaction mixture was cooled to room temperature. Evaporation of the DMF provided a solid mixture which was adsorbed onto silica gel (1 g). Purification by column chromatography (10% MeOH/CH_2Cl_2) provided porphyrin **61** as a dark red solid: UV-vis l_{max}= 458.5 nm; FAB MS m/z = 649 $[C_{32}H_{30}MnN_6O_6]^+$.

EXAMPLE 14

XVI. [5,15-Bis(dimethylamido)-10,20-bis(methoxycarbonyl)porphyrinato]-manganese(III) Chloride (63).



1. 5,15-Bis(dimethylamido)-10,20-bis(methoxycarbonyl)porphyrin (62).

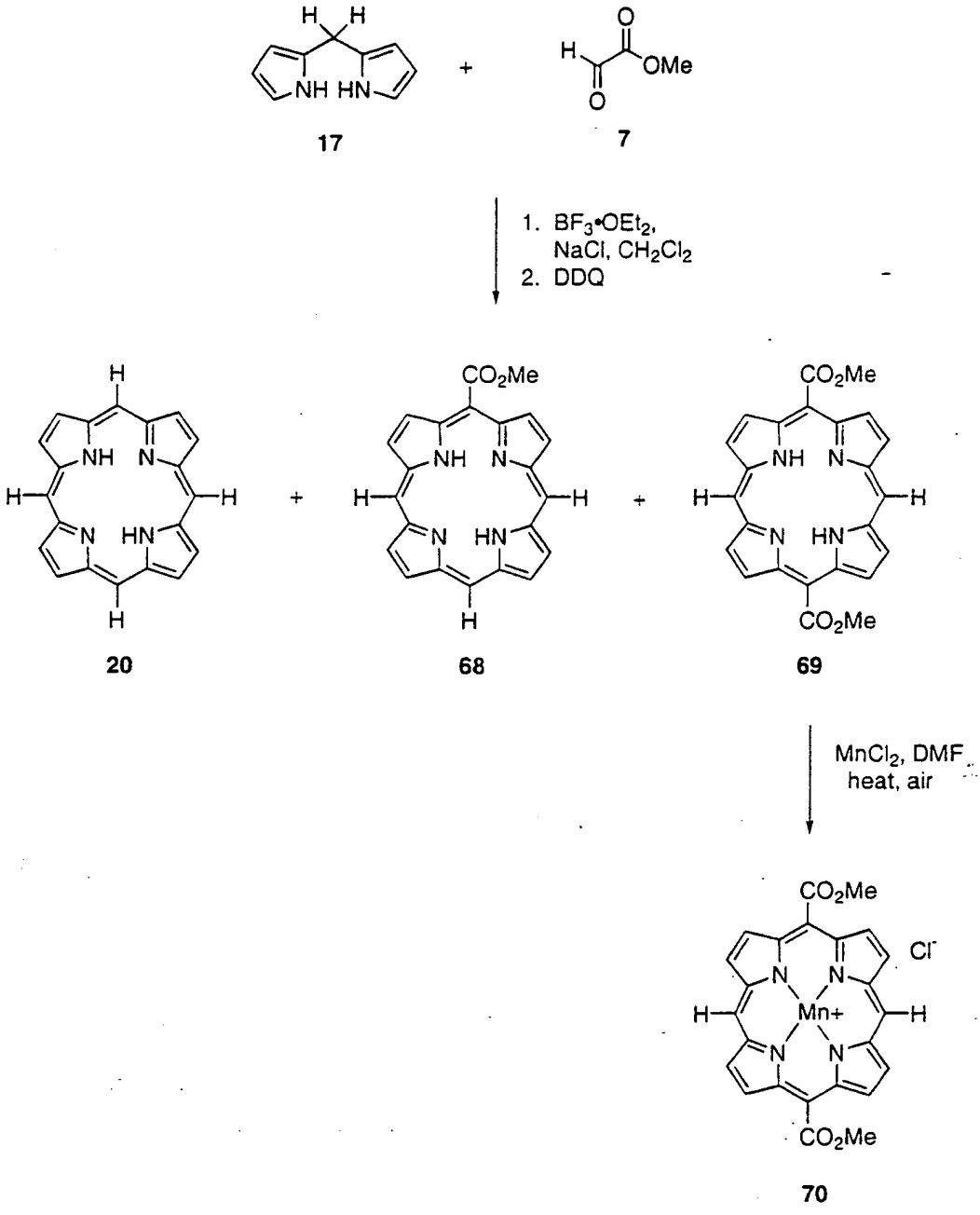
To a foil covered round-bottom flask equipped with a magnetic stirrer and a N_2 inlet was added consecutively dipyrromethane **59** (915 mg, 4.21 mmol), CH_2Cl_2 (400 μL), freshly distilled methyl glyoxylate (370 mg, 4.21 mmol) in CH_2Cl_2 (30 mL) and NaCl (25 mg, 0.43 mmol). The reaction mixture was stirred for 5-10 min, then BF_3OEt_2 (160 μL, 1.26 mmol) was added. After a 30 min stirring period, DDQ (1.43 g, 6.32 mmol) was added and the reaction mixture was stirred overnight. The solvent was removed *in vacuo* and the residue was adsorbed onto silica gel (4 g). Repeated purification by column chromatography (gradient elution with 0→10% CH_3OH/CH_2Cl_2) afforded porphyrin **62** (40 mg; 1.7%) as a dark violet solid mixture of atropisomers: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.1 (s, 2 H), 2.71, 2.78 (two singlets, 6 H), 3.81 (s, 6 H), 4.60 (s, 6 H), 9.30 (d, 4 H), 9.53 (d, 4 H); FAB MS m/z = 569 $[C_{30}H_{28}N_6O_6+H]^-$.

2. [5,15-Bis(dimethylamido)-10,20-bis(methoxycarbonyl)porphyrinato]-manganese(III) Chloride (63).

A solution of **62** (41 mg, 0.072 mmol) and $MnCl_2$ (181 mg, 1.44 mmol) in DMF (18 mL) was heated at 145°C for 1.5 h then exposed to a stream of air. The reaction mixture was heated for an additional 1 h then allowed to cool to room temperature overnight under a stream of air. Evaporation of the DMF provided a solid mixture which was adsorbed onto silica gel (1.5 g). Purification by column chromatography (gradient elution with 5→20% CH_3OH/CH_2Cl_2) provided a dark red solid. The solid was dissolved in CH_2Cl_2 (10 mL) and filtered through a fritted funnel. The residual solid was washed with 2% CH_3OH/CH_2Cl_2. The filtrates were combined and the solvent removed *in vacuo* to afford **63** (10 mg; 21%) as a black solid mixture of atropisomers: mp > 300°C; UV-vis λ_{max} =458.5 nm, ϵ = 3.33 x 10^4 L/cm-mol; FAB MS m/z = 621 $[C_{30}H_{26}MnN_6O_6]^+$.

EXAMPLE 15

XVIII. [5,15-Bis(methoxycarbonyl)porphyrinato]manganese(III) Chloride (70) and [5-(methoxycarbonyl)porphyrinato]manganese(III) Chloride (71).



1. 5-(Methoxycarbonyl)porphyrin (68) and 5,15-bis(methoxycarbonyl)-porphyrin (69)

In a foil covered 3 L round-bottom flask equipped with a magnetic stir bar and a N_2 inlet was added consecutively dipyrromethane **17** (2.92 g, 20 mmol), CH_2Cl_2 (1900 mL), methyl glyoxylate **7** (1.76 g, 20 mmol) as a solution in CH_2Cl_2 (100 mL) and NaCl (118 mg, 2 mmol). The reaction mixture was stirred for 5-10 min, then $BF_3{\cdot}OEt_2$ (740 μL, 6.0 mmol) was added. After a 30 min stirring period, DDQ (6.81 g, 30 mmol) was added and the solution was stirred for 2 d. The solvent was removed *in vacuo* producing a black tar, which was dissolved in CH_2Cl_2/MeOH (99:1) and filtered through a short plug of silica gel. The filtrate was concentrated, then adsorbed onto silica gel (1 g), and purified by column chromatography (gradient elution with 33% hexanes/CH_2Cl_2 →100% CH_2Cl_2) to provide porphyrins **20**, **68** (30 mg, 0.7%), and **69** (61 mg; 1.4%). Porphyrin **68**: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.6 (s, 2 H), 4.6 (s, 3 H), 9.49-9.53 (m, 6 H), 9.7 (d, 2 H), 10.3-10.5 (m, 3 H). Porphyrin **69**: ^{1}H NMR (300 MHz, $CDCl_3$) δ -3.2 (s, 2 H), 4.6 (s, 6 H), 9.5 (d, 4 H), 9.7 (d, 2 H), 10.4 (s, 2 H).

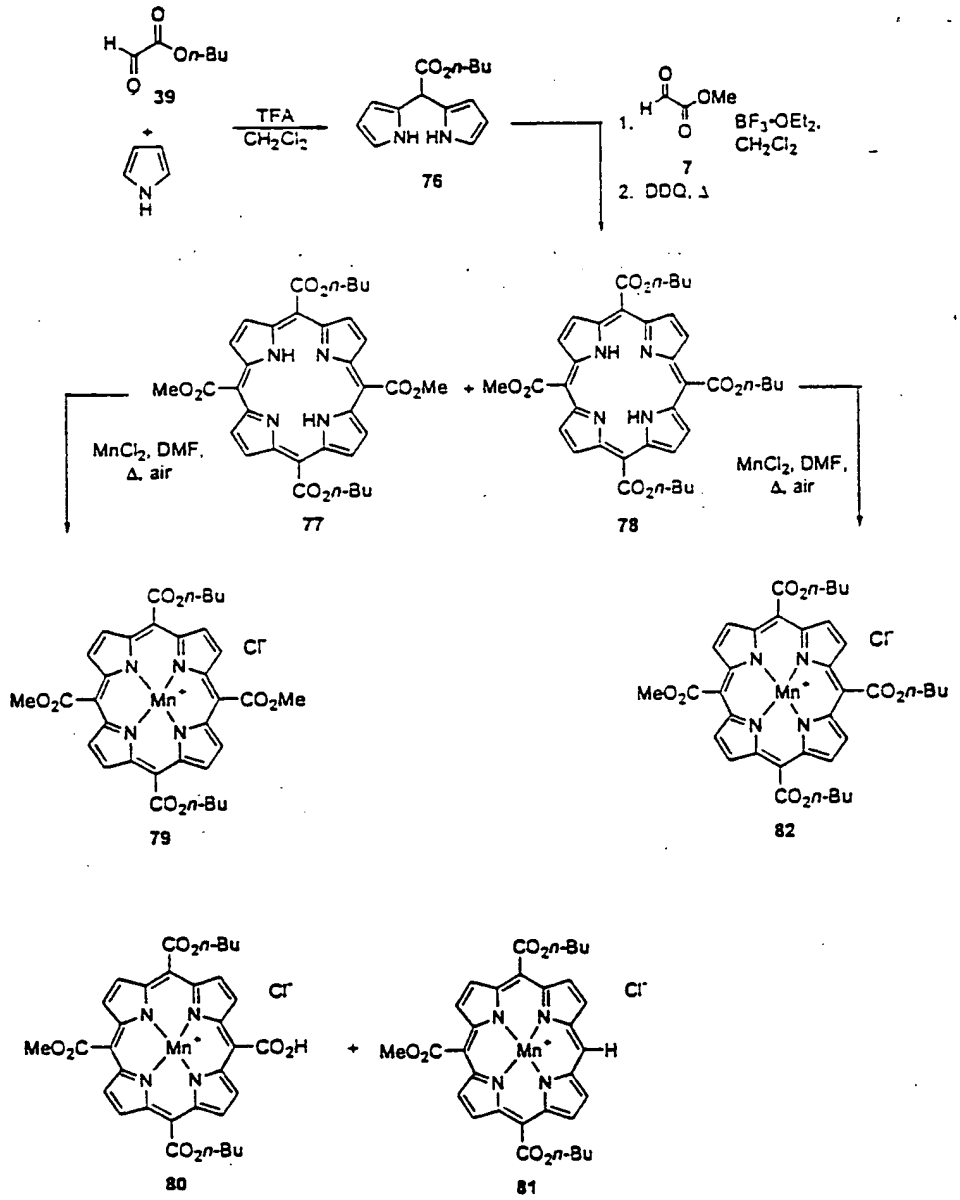
2. [5,15-Bis(methoxycarbonyl)porphyrinato]manganese(III) Chloride (70).

To a magnetically stirred solution of porphyrin **69** (61 mg, 0.142 mmol) in anhydrous DMF (70 mL) was added $MnCl_2$ (90 mg, 0.71 mmol). The reaction mixture was heated at 150 °C for 2 h then exposed to a stream of air. The reaction solution was heated for an additional 2-3 h, then the solution was allowed to cool to room temperature overnight under a stream of air. Then the reaction mixture was heated again to 150 °C and stirred for another 4 h. Evaporation of DMF *in vacuo* provided a solid mixture which was adsorbed onto silica gel (1 g). Purification by column chromatography (5%

MeOH/CH_2Cl_2) provided compound **70** (25 mg; 34%) as a dark solid: mp >300 °C; UV-vis λ_{max} = 452.0 nm, $\epsilon = 4.40 \times 10^4$ L/cm-mol; FAB MS m/z = 479 $[C_{24}H_{16}MnN_4O_4]^+$.

EXAMPLE 16

XX. [5,15-Bis(*n*-butoxycarbonyl)-10,20-bis(methoxycarbonyl)-porphyrinato]manganese(III) Chloride (79), [10,20-Bis(*n*-butoxycarbonyl)-5-carboxy-5-(methoxycarbonyl)porphyrinato]-manganese(III) Chloride (80), [5,15-Bis(*n*-butoxycarbonyl)-10-(methoxycarbonyl)porphyrinato]manganese(III) Chloride (81) and [5-methoxycarbonyl-10,15,20-tris(*n*-butoxycarbonyl)porphyrinato]-manganese(III) Chloride (82).



1. (*n*-Butoxycarbonyl)dipyrromethane (76).

A solution of *n*-butyl glyoxylate (**39**) (3.85 g, 29.6 mmol) in CH_2Cl_2 (100 ml) was magnetically stirred under N_2, in a flask covered with foil. Pyrrole (24.6 ml, 355 mmol) was added and the reaction mixture was allowed to stir overnight. A black product mixture was obtained after evaporation of solvents. This material was filtered through a plug of silica using CH_2Cl_2 as eluent. Column chromatography of the residue on silica gel (eluent 1:1 hexane: CH_2Cl_2) provided pure dipyrromethane **76** (1.04 g, 14%).

2. 5,15-Bis(*n*-butoxycarbonyl)-10,20-bis(methoxycarbonyl)porphyrin (77) and 5-Methoxycarbonyl-10,15,20-Tris(*n*-butoxycarbonyl)porphyrin (78).

In a foil covered, 500 mL three-neck round-bottomed flask equipped with a magnetic stirrer and a N_2 inlet was added consecutively methyl glyoxylate (376 mg, 4.27 mmol), dipyrromethane **39** (1.04 g, 4.23 mmol), NaCl (27 mg, 0.46 mmol), and CH_2Cl_2 (420 mL). The reaction mixture was stirred for 5-10 min then $BF_3 \bullet OEt_2$ (155 μL, 1.26 mmol) was added. After a stirring period of 30 min at room temperature, DDQ (1.43 g, 6.3 mmol) was added. The reaction mixture was stirred for an additional 2 h at room temperature, then the solvent was removed *in vacuo*. The residue was purified by repeated chromatographic purifications to provide porphyrins **77** (75 mg) and **78** (20 mg). Porphyrin **77**: ^{1}H NMR (300 MHz, $CDCl_3$) δ-3.41 (s, 2 H), 1.11 (t, 6H), 1.69 (m, 4 H), 2.12 (m, 4H), 4.59 (s, 6 H), 5.05 (t, 4 H), 9.49 (s, 8 H). Porphyrin **78**: ^{1}H NMR (300 MHz, $CDCl_3$)δ-3.34(s, 2 H), 1.11 (7, 9 H), 1.70 (m, 6 H), 2.13 (m, 6H), 4.60 (s, 3 H), 5.05 (t, 6 H), 9.51 (s, 8 H).

3. **[5,15-Bis(*n*-butoxycarbonyl)-10,20-bis(methoxycarbonyl)-porphyrinato]manganese(III) Chloride (79), [10,20-Bis(*n*-butoxycarbonyl)-5-carboxy-15-(methoxycarbonyl)porphyrinato]-manganese(III) Chloride (80), [5,15-Bis(*n*-butoxycarbonyl)-10-(methoxycarbonyl)porphyrinato]-manganese(III) Chloride (81).**

A solution of 77 (75 mg, 0.12 mmol) and $MnCl_2$ (163 mg, 1.30 mmol) in DMF (35 mL) was heated at 145°C for 2 h. The reaction mixture was then exposed to a stream of air as it cooled to room temperature. Evaporation of DMF provided a solid mixture which was absorbed onto 2 g silica gel. Purification by column chromatography (gradient elution with 0-7.5% MeOH/ CH_2Cl_2) provided porphyrin **79** (33 mg). Further purification of the remaining mixed fractions provided porphyrins **80** (10 mg), and **81** (1 mg). Porphyrin **79**: mp 200-205°C; UV-vis λ_{max} = 456.0 nm, ϵ = 9.50 x 10^4 L/cm-mol; FAB MS m/z = 679 $[C_{34}H_{32}MnN_4O_8]^+$. Porphyrin **80**: mp>300°C; UV-vis λ_{max} =460.0 nm, ϵ = 8.20 x 10^4 L/Cm-mol; FAB MS m/z = 665 $[C_{33}H_{30}MnN_4O_8]^+$. Porphyrin **81**: FAB MS m/z = 621 $[C_{32}H_{30}MnN_4O_6]^+$.

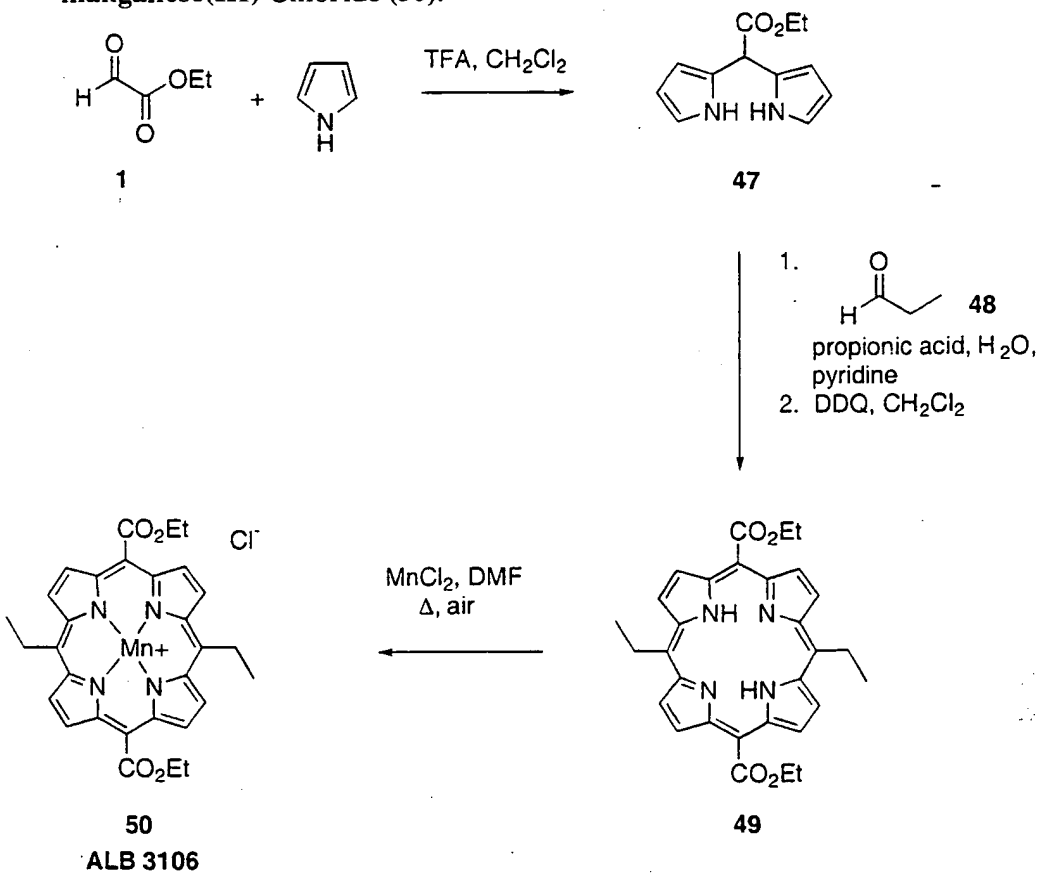
4. **[5-methoxycarbonyl-10,15,20-tris(*n*-butoxycarbonyl)porphyrinato]-manganese(III) Chloride (82).**

A solution of **78** (20 mg, 0.03 mmol) and $MnCl_2$ (28 mg, 0.22 mmol) in DMF (25 mL) was heated at 140°C for 1.5 h. Additional $MnCl_2$ (26 mg, 0.21 mmol) was added to the reaction mixture. The reaction mixture was exposed to a stream of air and heating was continued for an additional 4 h. The reaction mixture was cooled to room temperature overnight under a stream of air. Evaporation of DMF provided a solid mixture, which was adsorbed onto 2 g silica gel. Purification by column chromatography (gradient elution with 2-7.5% MeOH/CH_2Cl_2) provided porphyrin **82** (6 mg): mp 180-185°C; UV-vis

λ_{max} = 456.0 nm, $\epsilon = 7.10 \times 10^4$ L/cm-mol; FAB MS m/z = 721 $[C_{37}H_{38}MnN_4O_8]^+$.

EXAMPLE 17

XII. [5,15-Bis(ethoxycarbonyl)-10,20-bis(ethyl)porphyrinato]-manganese(III) Chloride (50).



1. *meso*-(Ethoxycarbonyl)dipyrromethane (47).

In a round-bottom flask equipped with a magnetic stir bar and N_2 inlet was placed ethyl glyoxylate (12.6 g, 0.123 mol), pyrrole (102 mL, 1.48 mol) and CH_2Cl_2 (700 mL). Trifluoroacetic acid (3.8 mL, 0.049 mol) was then added. The resulting dark solution was stirred overnight at room temperature,

transferred into a separatory funnel, diluted with CH_2Cl_2, then washed with H_2O, saturated aqueous $NaHCO_3$, H_2O and brine. The organic portion was dried (Na_2SO_4), filtered and the solvent removed *in vacuo*. The crude product was repeatedly chromatographed on silica gel (elution with 50% CH_2Cl_2/hexanes). Recrystallization from CH_2Cl_2/hexane provided the product **47** as white crystals (9.39 g; 35%): mp 70-75 °C; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.31 (t, 3 H), 4.24 (q, 2 H), 5.10 (s, 1 H), 6.09 (broad s, 2 H), 6.16 (m, 2 H), 6.72 (broad s, 2 H), 8.45 (broad s, 2 H).

2. 5,15-Bis(ethoxycarbonyl)-10,20-bis(ethyl)porphyrin (49).

In a foil-covered flask fitted with an air condensor, dipyrromethane **47** (150 mg, 0.687 mmol) was magnetically stirred in propionic acid (5 mL), H_2O (0.2 mL) and pyridine (17 μL) at 90 °C for 5 min (Neya, S.; Funasaki, N. *J. Heterocyclic Chem.* 1997, *34*, 689-690). Propionaldehyde (25 μL, 0.34 mmol) was added and the reaction mixture was stirred for 40 min. Another portion of propionaldehyde (10 μL, 0.14 mmol) was added to the reaction mixture, stirred for 2 h and diluted with CH_2Cl_2. The organic phase was washed with H_2O, 0.05 N NaOH (2 x), and H_2O, dried (Na_2SO_4), filtered and the solvent was removed *in vacuo*. The residue was dissolved in CH_2Cl_2 (250 mL) and DDQ (108 mg, 0.48 mmol) was added. After stirring overnight, the sample was adsorbed onto silica gel (4 g). Purification by column chromatography (gradient elution 50→100% CH_2Cl_2/hexane) provided porphyrin **46** (12 mg; 10%) as a dark solid: ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.81 (t, 6 H), 2.13 (t, 6 H), 5.11-4.99 (m, 8 H), 9.44 (d, 4 H), 9.54 (d, 4 H); FAB MS m/z = 511 $[C_{30}H_{30}N_4O_4 + H]^+$.

3. [5,15-Bis(ethoxycarbonyl)-10,20-bis(ethyl)porphyrinato]manganese(III) Chloride (50).

To a magnetically stirred solution of porphyrin **49** (12 mg, 0.025 mmol) in anhydrous DMF (4 mL) was added $MnCl_2$ (23 mg, 0.19 mmol). The reaction

flask was fitted with a reflux condensor and the solution was heated to 125 °C; air was then pumped into the reaction vessel, and the solution was stirred overnight. The flask was allowed to cool to room temperature, then the solvent was removed *in vacuo*. The crude material was adsorbed onto silica gel (1 g) and chromatography on silica gel (gradient elution 0→6% MeOH/CH_2Cl_2) provided **47** (3.5 mg; 24%) as a dark solid: mp >300 °C; UV-vis λ_{max} = 462.5 nm, $\varepsilon = 3.43 \times 10^4$ L/cm-mole; FAB MS m/z = 563 $[C_{30}H_{28}MnN_4O_4]^+$.

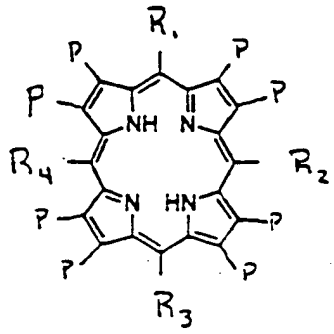
EXAMPLE 18

ATTENUATION OF HYPEROXIC LUNG INJURY BY A NOVEL CATALYTIC ANTIOXIDANT

Pulmonary toxicity due to hyperoxia is thought to be related to the formation of reactive oxygen species, including superoxide. Increased levels of antioxidant enzymes, such as superoxide dismutases (SOD) and catalase, have been associated with increased survival and adaptation to hyperoxia in rats and mice. The role of a catalytic antioxidant in conferring protection from hyperoxic lung injury to rats was examined. The compound is a manganic porphyrin (AEOL-11201 (see Fig. 1)) with a broad spectrum of antioxidant properties. Male Sprague-Dawley rats were exposed to 100% O_2, 635 mmHG, for 7 days. The animals were injected with the compound at 15 mg/kg, or the vehicle intraperitoneally every 24 hours. Perivascular edema, a marker of hyperoxic lung injury, was evaluated on hematoxylin and eosin stained lung sections. Compared to the air control animals, the oxygen exposed group developed significant perivascular edema. AEOL-11201 significantly reduced the edema of small to medium sized vessels in O_2 exposed rats. These results indicate that manganic porphyrins are useful as therapeutic antioxidants in disease states in which reactive oxygen species are involved.

WHAT IS CLAIMED IS:

1. A compound of formula



I

or pharmaceutically acceptable salt thereof,

wherein

R_1 and R_3 are, independently:

$-CO_2C_{1-4}$ alkyl; or

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3;

R_2 is:

-H

$-C_{1-4}$alkyl

-COOH

$-CO_2C_{1-4}$ alkyl,

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

$-CON(CH_3)_2$, or

$-CX_3$, wherein X is halogen; and

R_4 is:

-H,

$-C_{1-4}$alkyl

-COOH,

$-CO_2C_{1-4}$ alkyl,

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

$-CON(CH_3)_2$, or

$-CX_3$, wherein X is halogen, and

each P is, independently, an electron withdrawing group or hydrogen.

2. The compound according to claim 1 wherein R_1 and R_3 are, independently, $-CO_2C_{1-4}$alkyl or $-CO_2CH_2CX_3$, R_2 is -H, $-CO_2C_{1-3}$alkyl, $-CO_2CH_2CX_3$, $-CON(CH_3)_2$ or CX_3 and R_4 is -H, -COOH, $-CO_2C_{1-3}$alkyl, $-CON(CH_3)_2$ or $-CX_3$.

3. The compound according to claim 2 wherein R_1 and R_3 are, independently, $-CO_2C_{1-3}$alkyl, R_2 is $-CO_2C_{1-3}$alkyl, $-CON(CH_3)_2$ or $-CX_3$, and R_4 is -H, -COOH, $-CO_2C_{1-3}$alkyl, $-CON(CH_3)_2$ or $-CX_3$.

4. The compound according to claim 3 wherein R_1 or R_3 is $-CO_2CH_3$, $-CO_2CH_2CH_3$, or $-CON(CH_3)_2$, R_2 is $-CO_2CH_3$, $-CO_2CH_2CH_3$, or CX_3, and R_4 is -H, -COOH, $-CO_2CH_3$, $-CO_2CH_2CH_3$ or CX_3.

5. The compound according to claim 3 wherein R_1, R_2 and R_3 are, independently, $-CO_2CH_3$ or $-CO_2CH_2CH_3$, and R_4 is -H, -COOH, $-CO_2CH_3$ or

$-CO_2CH_2CH_3$ or $-CO_2CH_2CH_3$.

6. The compound according to claim 5 wherein R_1, R_2, R_3 and R_4 are, independently, $-CO_2CH_3$ or $-CO_2CH_2CH_3$.

7. The compound according to claim 1 wherein each P is, independently, hydrogen or an electron withdrawing group selected from the group consisting of $-NO_2$, a halogen, a nitrile, a vinyl group and a formyl group.

8. The compound according to claim 1 wherein at least one P is a halogen.

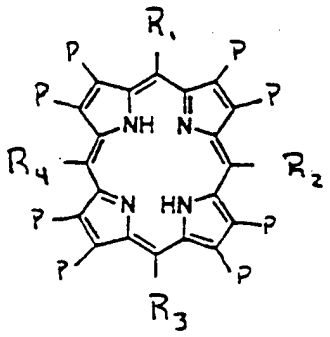
9. The compound according to claim 1 wherein each P is hydrogen.

10. The compound according to any one of claims 1-9 wherein said compound is complexed with a metal selected from the group consisting of manganese, iron, copper, cobalt, nickel and zinc.

11. The compound according to claim 10 wherein said compound is complexed with manganese.

12. The compound according to claim 1 wherein R_1, R_2, R_3 and R_4 are, independently, $-CO_2CH_3$ or $-CO_2CH_2CH_3$, each P is a hydrogen, and said compound is complexed with manganese.

13. A method of protecting cells from oxidant- induced toxicity comprising contacting said cells with a protective amount of a compound of formula



I

or pharmaceutically acceptable salt thereof,

wherein

R_1 and R_3 are, independently:

$-CO_2C_{1-4}$ alkyl; or

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3;

R_2 is:

-H

$-C_{1-4}$alkyl

-COOH

$-CO_2C_{1-4}$ alkyl,

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

$-CON(CH_3)_2$, or

$-CX_3$, wherein X is halogen; and

R_4 is:

-H,

$-C_{1-4}$alkyl

-COOH,

$-CO_2C_{1-4}$ alkyl,

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

$-CON(CH_3)_2$, or

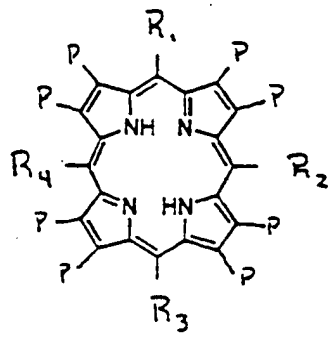
$-CX_3$, wherein X is halogen, and

each P is, independently, an electron withdrawing group or hydrogen.

14. The method according to claim 13 wherein said compound is complexed with a metal selected from the group consisting of manganese, iron, copper, cobalt, nickel or zinc.

15. The method according to claim 13 wherein said cells are mammalian cells.

16. A method of treating a patient suffering from a condition that results from or that is exacerbated by oxidant-induced toxicity comprising administering to said patient an effective amount of a compound of formula



I

or pharmaceutically acceptable salt thereof,

wherein

R_1 and R_3 are, independently:

$-CO_2C_{1-4}$ alkyl; or

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3;

R_2 is:

-H

$-C_{1-4}$alkyl

-COOH

$-CO_2C_{1-4}$ alkyl,

-$CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

-$CON(CH_3)_2$, or

-CX_3, wherein X is halogen; and

R_4 is:

-H,

-C_{1-4}alkyl

-COOH,

-CO_2C_{1-4} alkyl,

-$CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

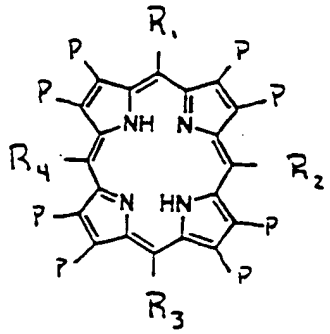
-$CON(CH_3)_2$, or

-CX_3, wherein X is halogen, and

each P is, independently, an electron withdrawing group or hydrogen.

17. The method according to claim 16 wherein said compound is complexed with a metal selected from the group consisting of manganese, iron, copper, cobalt, nickel or zinc.

18. A method of treating a pathological condition of a patient resulting from degradation of NO· or a biologically active form thereof, comprising administering to said patient an effective amount of a compound of formula



I

or pharmaceutically acceptable salt thereof,

wherein

R_1 and R_3 are, independently:

$-CO_2C_{1-4}$ alkyl; or

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3;

R_2 is:

-H

$-C_{1-4}$alkyl

-COOH

$-CO_2C_{1-4}$ alkyl,

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

$-CON(CH_3)_2$, or

$-CX_3$, wherein X is halogen; and

R_4 is:

-H,

$-C_{1-4}$alkyl

-COOH,

$-CO_2C_{1-4}$ alkyl,

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

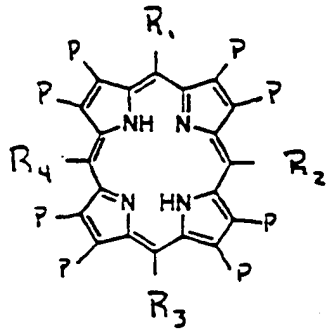
$-CON(CH_3)_2$, or

$-CX_3$, wherein X is halogen, and

each P is, independently, an electron withdrawing group or hydrogen.

19. The method according to claim 18 wherein said compound is complexed with a metal selected from the group consisting of manganese, iron, copper, cobalt, nickel or zinc.

20. A method of treating a patient for an inflammatory disease comprising administering to said patient an effective amount of a compound of formula



I

or pharmaceutically acceptable salt thereof,

wherein

R_1 and R_3 are, independently:

$-CO_2C_{1-4}$ alkyl; or

$-CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3;

R_2 is:

-H

$-C_{1-4}$alkyl

-COOH

$-CO_2C_{1-4}$ alkyl,

-$CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

-$CON(CH_3)_2$, or

-CX_3, wherein X is halogen; and

R_4 is:

-H,

-C_{1-4}alkyl

-COOH,

-CO_2C_{1-4} alkyl,

-$CO_2(CH_2)_nCX_3$, wherein X is halogen and n = 1 to 3,

-$CON(CH_3)_2$, or

-CX_3, wherein X is halogen, and

each P is, independently, an electron withdrawing group or hydrogen.

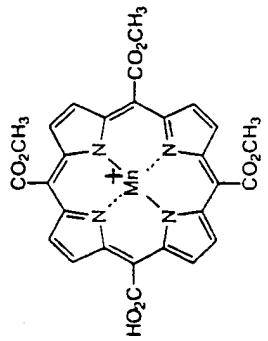
21. The method according to claim 20 wherein said compound is complexed with a metal selected from the group consisting of manganese, iron, copper, cobalt, nickel or zinc.

22. The method according to claim 21 wherein said metal is manganese.

23. The method according to claim 20 wherein said inflammatory disease is an inflammatory lung disease.

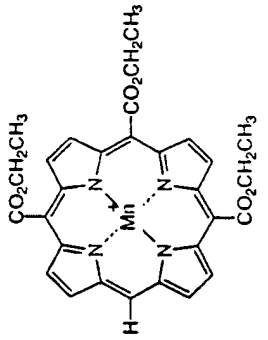
24. The method according to claim 23 wherein said inflammatory lung disease is bronchopulmonary disease.

25. The method according to claim 23 wherein said inflammatory lung disease is asthma.



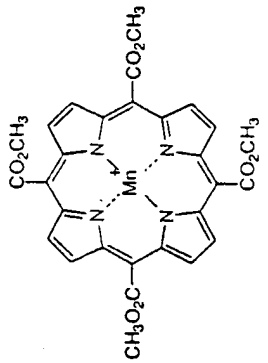
AEOL-11201

SOD 13.2
TBARS 0.2



AEOL-11202

X-
0.1

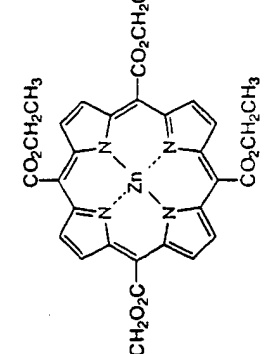


AEOL-11203

X-
0.05

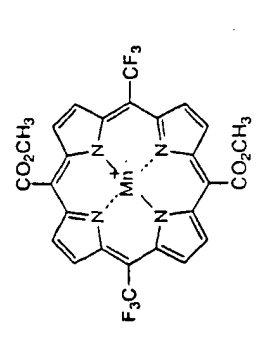
AEOL-11204

X-
0.3



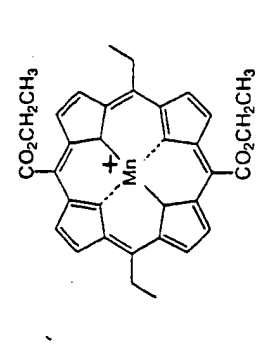
AEOL-11205

SOD X-
TBARS 1.3



AEOL-11206

X-
>90



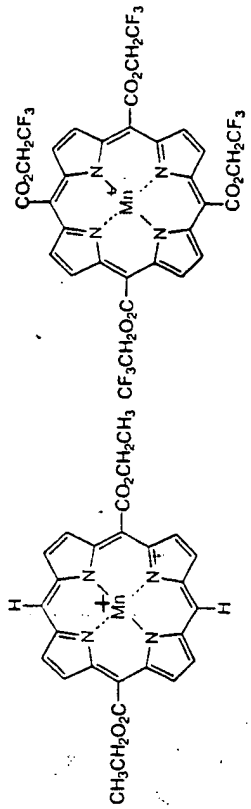
AEOL-11207

interfers
0.2

AEOL-11212

interfers
14.7

Fig. 1

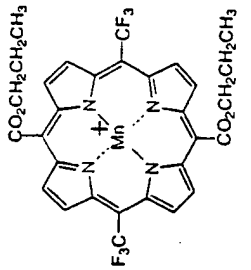


AEOL-11218

SOD 4
TBARS 15

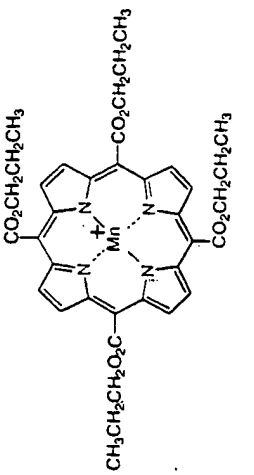
AEOL-11217

SOD interfers
TBARS 9.9



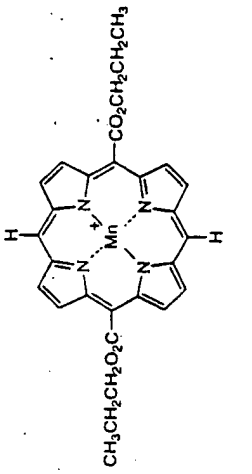
AEOL-11216

SOD 98
TBARS 4



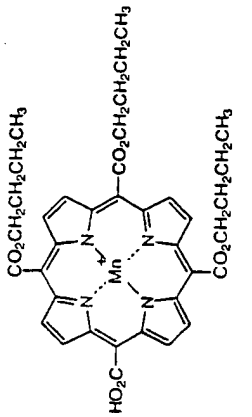
AEOL-11215

SOD 221
TBARS 3.7



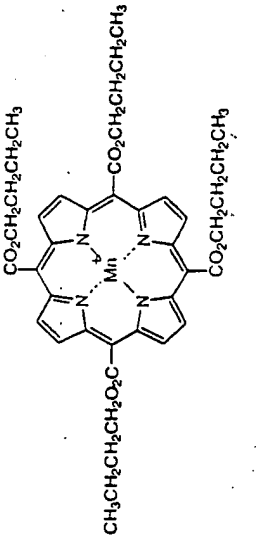
AEOL-11221

SOD interfers
TBARS 8.4



AEOL-11220

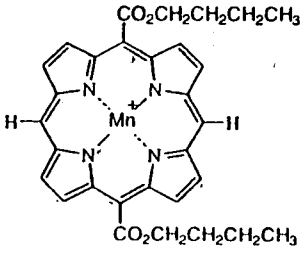
SOD interfers
TBARS 21



AEOL-11219

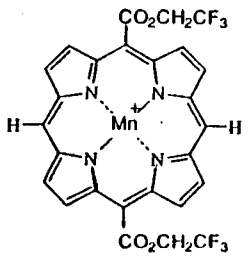
SOD interfers
TBARS 8.2

Fig. 1 (continued)



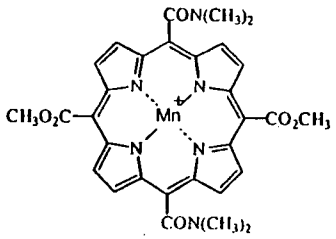
AEOL-11223

SOD interfers
TBARS 4.4



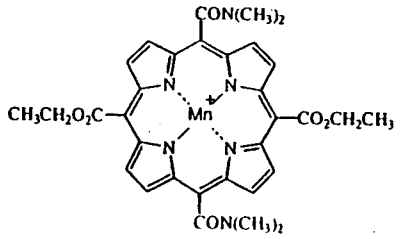
AEOL-11225

interfers
4.4



AEOL-11229

0.7
2.1



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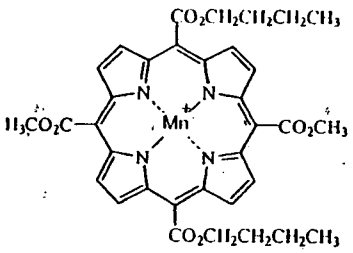
5.6
1.5



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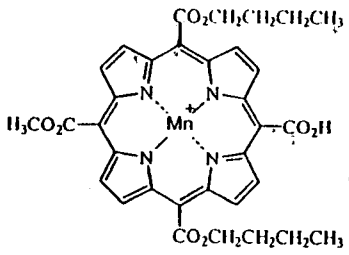

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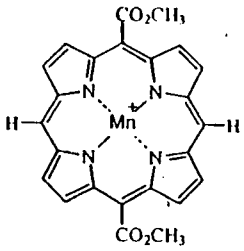

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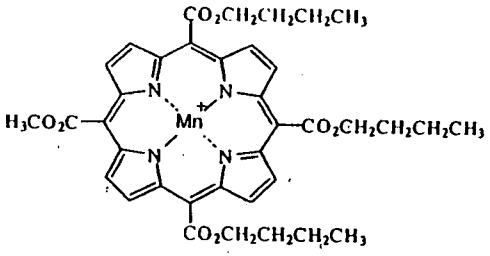

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ND
8.6

Fig. 1 (continued)

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US99/08905

A. CLASSIFICATION OF SUBJECT MATTER
IPC(6) :A61K 51/04; C07D 487/22
US CL :514/184,183; 540/145
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 514/184,183; 540/145

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 5,674,467 A (MAIER et al.) 07 October 1997, col. 2, lines 30-50.	1-12
X	US 5,599,924 A (THERIEN et al) 04 February 1997, col. 2, lines 5-35.	1-12

☐ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

*	Special categories of cited documents:	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"A"	document defining the general state of the art which is not considered to be of particular relevance	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"E"	earlier document published on or after the international filing date	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"L"	document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"&"	document member of the same patent family
"O"	document referring to an oral disclosure, use, exhibition or other means		
"P"	document published prior to the international filing date but later than the priority date claimed		

Date of the actual completion of the international search 06 JULY 1999	Date of mailing of the international search report 19 JUL 1999
Name and mailing address of the ISA/US Commissioner of Patents and Trademarks Box PCT Washington, D.C. 20231 Facsimile No. (703) 305-3230	Authorized officer P. Lawrence for PKSRIPADA Telephone No. (703) 308-1235

Form PCT/ISA/210 (second sheet)(July 1992)*